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SOME PHYSICO-CHEMICAL ASPECTS OF LIFE AND EVOLUTION IN RELATION TO THE LIVING STATE

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INTRODUCTION

IN this paper a treatment of several matters of basic significance to theoretical biology is undertaken from a physico-chemical standpoint. Purposive interpretations are avoided, consistently with contemporary efforts to establish an operational basis for the application of the physico-chemical properties of matter to the analysis and explication of biological phenomena. The author (1945) has presented a brief introductory treatment of the topic of causal factors in the origin of life, and this topic is considered here in greater detail.

I. THE NATURE OF LIFE

We shall first consider certain theoretical aspects of living entities in so far as they may provide an indication of the probable nature of the physical factors to which the existence of life may be attributed. Thus, it is important for us to determine what material condition is basically necessary (though not sufficient) in order that the phenomena of the living state can be manifested. In adopting this program we are assuming that the properties of the living state are able to make their appearance because certain physico-chemical relationships can attain greater or new expression in a system characterized by the necessary material conditions.

Reproductive Aspects. There are two properties of life which have received the greatest amount of attention from investigators seeking to identify the fundamental life-property. These are reproductivity and organization. We shall briefly discuss these two important life-properties, for the consideration is useful in indicating the nature of the material conditions required in order that the living state may be attained. Considering the former life-property, a superficial examination is sufficient to reveal that one aspect, the potentiality of producing progeny, can not be considered the fundamental property of life. We conclude this because living organisms can be treated in such a manner that production of progeny is not even potentially possible, while other vital functions are essentially unchanged. Thus, we must consider reproductivity as it pertains to growth, *i.e.*, the reproduction of the various constituents of the organism. If we regard it in this light, and as fundamental, it is necessary to conclude that no matter how simple a form of life may be, reproduction of its constituent units is always the primary vital process. That is to say, that even in its most primitive form, life must consist of reproductive or self-duplicating units.

Similar considerations have led a number of investigators to propose that the simplest forms of life are complex self-duplicating molecules (Alexander, 1928, and Troland, 1914, 1916, 1917) and that it is quite in order to speak of a living molecule (Vaughan, 1927). Alexander states, "We should expect a cellular type to arise before a multicellular, a nuclear before a cellular, a chromosomic before a nuclear, a genic before a chromosomic, and, hypothetically first of all, the genetomolecular type." There is significant evidence, however, to support only the first and fourth of Alexander's expectations, while the other three are based upon assumptions about the functional relationships of the elements of the cell and nucleus that are not permissible at the present state of our knowledge of these relationships.

The arguments for the "living molecule" hypothesis are mainly supported by the evidence of the properties of enzymes and viruses. In the case of viruses, however, it has not yet been proved conclusively that they are pure nucleoprotein in constitution. There is some direct and indirect evidence to support the contention that small amounts of non-nucleoprotein constituents are also present (see Bawden's book, 1939, for material of this sort). The indirect evidence centers about the fact that from a biological standpoint the properties of various viruses are in many cases so sharply defined and differentiated from one another (*in vivo* and *vitro*) that the view that they are all merely different combinations of proteins and nucleic acids appears to be inadequate. Yet, even assuming that viruses are pure nucleoprotein, we can not consider them to be primordial forms of life as such, for they can only thrive in the manner of parasites in living tissue and they have not been known to grow, metabolize or reproduce *in vitro*.

A great shortcoming of the "living molecule" theory is that it fails to take into account the endothermal nature of the processes resulting in the formation of nucleoproteins and the properties of these substances. Only those constituents of living organisms which contain nucleic acid have been observed to grow and/or reproduce directly by self-duplicating processes (Claude, 1943). Because of the great complexity and specificity of the molecules formed and the fact that the free energy change is positive (and thus the "autosynthetic" reaction alone is not spontaneous), these processes are dependent upon the organizational nature and energy-yielding reactions characterizing protoplasm itself. *Protein-type substances are very unstable under non-protoplasmic conditions and extremely unstable in dilute solution.* Perhaps the most characteristic property of the soluble proteins is the ease with which they undergo denaturation (Conant, 1942). Thus, the formation or survival of protein-type substances under nonprotoplasmic conditions is highly improbable.

Arguments based upon enzyme activity can support the "Living Molecule" hypothesis in only a superficial manner. In precursor activation by proteolytic enzymes the enzyme produced by the activation of the precursor is determined by the precursor itself and not only by the enzyme used for activation. Thus, if trypsin is added to a solution of chymotrypsinogen, this is converted into chymotrypsin, not trypsin. Of greater significance in our consideration is the fact that the autocatalysis of an enzyme from its precursor is not a *synthetic* process but very likely merely an hydrolysis (Northrop, 1938). The reactions of nucleoprotein synthesis *in vivo* are characterized by great increases in free energy, whereas precursor activation involves comparatively negligible free energy changes and the process is degradative.

In view of the above considerations, the "living molecule" hypothesis is no longer tenable. The view of reproductivity being the fundamental property of life must be regarded in the same light, for it inevitably reduces to the "living molecule" hypothesis. *It is also evident that reproductivity as it pertains to the living state is dependent upon and inseparable from protoplasmic organization.*

Organizational Aspects. There are many investigators who adhere to the proposition that organization is the fundamental life-property, among them Calkins (1926), Conklin (1940), Haldane (1931), Oparin (1938) and Osborn (1917). It is unnecessary to discuss organization in detail for, with respect to the fundamental life-property, it is generally regarded as the only alternative to reproductivity and we have already concluded that this latter property is dependent upon protoplasmic organization.

When we speak of the organization of protoplasm we have in mind specific functions with respect to which it is organized. These functions are the ones observed to be characteristic of the living state, namely, the conversion and storage of energy, growth and reproduction, maintenance of structure and form, etc. However, be-

cause of the purpose of our program, we are more concerned with the material conditions which protoplasmic organization requires than we are with the character of this organization.

Systemic Basis. Protoplasmic organization is a complex state of the organization of matter into highly integrated systems, and it is only possible upon or above the level of a system of molecular (and infra-molecular) systems.¹

Before proceeding, let us reflect upon the fact that the objective of our consideration of the material condition required in order that the living state may be attained is what this consideration may lead us to conclude about the nature of the physical factors operative in the origin of life. For this reason, we must expect to treat of entities in a state characterized by this condition, and perhaps possessing some, but not all, of the properties of life. We are not assuming that there is any sharply defined material level at which systems of matter assume the properties of life. Rather we are allowing for, and anticipating, the probability that under certain conditions systems of matter "evolve" in such a manner that they progressively give expression to material characteristics which are only latent or feebly expressed in less complex material states. Then, after they have reached some arbitrary point by a relatively continuous process, *we* simply define them as being alive. Thus, we do not assume that functional properties manifested by life are necessarily unique to the living state or special aspects of material relationships possible *only* in living systems. We do consider that, perhaps, *life is a material entity possessing a certain combination of systemic characteristics, the group of which as a whole is manifested to a great degree by living systems (and certain of which may have not thus far been detected in non-living sys-*

¹ A "system of molecular systems" denotes a system consisting of many molecular species. Consistent with this terminology a molecular system is a system consisting of a single molecular species.

tems), but none of which are necessarily intrinsic to living systems alone.

Schrödinger (1945) has quite clearly pointed out that living systems are likely to involve "laws of physics" which are hitherto unknown, but which, when revealed, will form just as integral a part of the science as those already recognized. He further suggests that there is not likely to be any "new force" directing the behavior of the single atoms within a living system, and that if such a force appears to exist, it is only because the construction of the system is different from anything we have yet tested in the physical laboratory. As we have concluded, this does not imply that such relationships as may have appeared only in living systems thus far are intrinsic to them alone, and that they could not exist either in other types of naturally occurring systems or in "test-tube" systems.

We may assign a level of complexity to the systemic character which appears to be the material condition required in order that the living state may be attained. Thus (as above) it may be termed as being upon the level of a system of molecular systems. Perhaps it ought to be placed upon a more complex material level, but, since the higher levels are so vaguely defined at present, there does not appear to be any significant advantage in so doing.

Many of the phenomena of the vital processes of life are clearly characteristic of a system of molecular systems. For example, the phenomena of the colloidal state, which are of great importance, can not be manifested at a lower material level. The same may be said of the interphase membrane-type phenomena bounding organismal units. These matters are further considered in the section on the origin and nature of primordial systems.

The preceding considerations are useful in that they lead us to the reasonable conclusion that *the causal factors in the origin of living systems are, at the lowest level, resolvable into those factors which favor the association of systems of molecular systems.*

II. THE ORIGIN OF LIFE

Spatial Distribution and the Influence of the Sun. Passing now to the consideration of the origin of life let us first consider the spatial distribution of living organisms. It is a prominent feature of living systems that they are found to occur only over the surface of the earth or in a shallow outer layer. This fact indicates that the origin of life is in some manner related to physico-chemical factors which are maximally operative at the surface of the earth.

The fact that surface matter is exposed directly to the radiant energy (in highly energized quanta) emitted by the sun appears to be a most important distinguishing characteristic.² The state of matter occurring at the surface of the earth is determined not only by internal constitutional characteristics but also this afferent energy. For this reason the physico-chemical state of surface matter differs from that of matter similarly constituted but not receiving energy from an external source. The receipt of energy from the sun, or an equivalent agent, is a necessary condition to the continued existence of life. Although sunlight has an attenuating effect on most forms of bacteria, they are dependent, nevertheless, upon energy from the sun for their continued existence. Life, on the earth, then, is *ultimately* dependent upon the effects of solar radiation, and forms not utilizing this energy "directly" can only continue to live through direct or indirect contact with forms that do.

Carbon and Nitrogen. Constitutionally and functionally the elements carbon and nitrogen play very important roles in living systems, and the former element is regarded as the more important of the two. With re-

² There is considerable evidence against free atmospheric oxygen being an important surface factor in the origin of life. Oparin (1938) has presented convincing arguments that oxygen is beyond a doubt a secondary component of our atmosphere resulting from the activity of living organisms. It is also believed that the presence of carbon dioxide and nitrogen in the earth's atmosphere is of secondary biological origin (see Oparin's book for literature references and an excellent discussion of these topics).

spect to their insignificant quantitative appearance in the earth's crust these two elements occur in entirely disproportionate amounts in living organisms (Lotka, 1925). For these reasons it is important to investigate the properties of these elements to which their outstanding role in living systems may be attributed.

The element carbon is distinguished by the fact that it is relatively electroneutral. This is a consequence of the nature of its valence shell of electrons, in which respect it occupies a position intermediate between highly electropositive and electronegative elements. Thus, occupying an electron valence shell group midway between the stable electron states of helium and neon, carbon has relatively little tendency either to lose or gain electrons (Fieser and Fieser, 1944). In this quality of being fundamentally neutral carbon occupies a unique place even among Group IV elements. This is because the valence electrons of carbon are in the "L" shell, very close to the nucleus, and thus are held closely to the atomic sphere. In the other Group IV elements the valence electrons are farther away from the positive nucleus so that the electronic binding energy is less. These properties of the carbon shell of valence electrons are the basis for its characteristic electron sharing process and the extraordinary capacity of carbon atoms to bond with one another and to form atomic associations. The exceptional lability of carbon compounds, *e.g.*, the overwhelming variety of chemical transformations which hydrocarbons and their derivatives are capable of entering into and the readiness with which unsaturated carbon compounds bond with some other atoms, is likewise attributable to the properties of the carbon shell of valence electrons. The formation of bonds between carbon and other elements is characterized by relatively lesser energies of formation than are generally involved when more electrovalent atoms bond. On the electronegativity scale of elements Pauling (1939) assigns an electronegativity value to carbon which is midway between the values for lithium and fluorine.

Nitrogen also possesses very distinctive properties. Molecules of this element are characterized by comparative inertness. Thus, at ordinary temperatures nitrogen is comparatively indifferent to, and does not readily combine with, other elements. The energies of formation of bonds of nitrogen with other atoms are in general relatively low compared to those of bonds between other elements, and the N-N bond itself possesses a very low energy of formation. For this reason nitrogen-containing compounds are comparatively unstable and active. These properties also find their basis in the nature of the nitrogen valence shell of electrons.

Thus, we find that the properties of both carbon and nitrogen are such that, in a material aggregate, compounds in which these elements occur are much more likely to enter into reactions characterized or initiated by relatively small increments in energy than are the more stable compounds consisting of more electrovalent atoms.

Physico-chemical Aspects. These factors indicate that thermodynamic considerations may be of great importance in understanding the interactions of matter which lead to the formation of living systems. Let us, then, consider certain physico-chemical aspects of the earth-system from the standpoint of the following considerations. A specified system in any given environment tends to reach an equilibrium state characterized by a maximum in the entropy of the system and its environment. Otherwise stated, systems tend to reach conditions of maximum probability with respect to their environment and internal constitution. The most stable state is, by definition, the most probable state. Because of the rotation of the earth, such an equilibrium state characterizing either of the day-night temperature limits, if reached, is not maintained. Instead the energy and entropy of matter at the earth's surface oscillate from day to night as energy is absorbed and emitted. It is important to consider, then, the manner in which the components of the earth's surface may have reacted because of this systematic oscillation factor. Since it is

a characteristic of material systems that the above consideration applies to them, let us see how this characteristic might resolve itself in an imposed systematic temperature oscillation condition which tends to favor endothermal reactions during one period and exothermal reactions during another.

The absorption of energy by a system can cause two essentially different effects. The specific effect depends chiefly upon the chemical constitution and temperature of the system and the amount of and nature of the afferent energy. If, for example, the system consists only of aggregates of highly stable molecules (such as those formed between elements of Group I and Group VII) at normal temperatures encountered upon the earth, the addition of amounts of energy comparable to that received from the sun will merely result in an increase in the temperature of the system. On the other hand, if the system also contains labile molecules, *i.e.*, molecules the formation and decomposition of which are caused or accompanied by small changes in energy, the result of the addition of this energy will be twofold. One result will be to raise the temperature of the system, as noted above, while a second will be to bring about the occurrence of endothermal reactions with a resultant increase in the potential energy of the system.

Since we are concerned with factors operative in the origin of life, we need not consider an upper temperature limit in excess of the boiling point of water. Study of the properties of life indicates very strongly that the living state is not compatible with temperatures above this point. With such an upper limit, and in view of the amount of the afferent energy received per unit area of the earth per rotation, it is evident that the temperature oscillation through the period under consideration could have an appreciable effect only upon molecules whose formation and dissociation reactions involve relatively small energy changes. Thus, the carbon-nitrogen type compounds³ may be assumed to have been continually in-

³ Compounds in which carbon or nitrogen or both figure prominently.

volved in processes of formation and decomposition as the temperature oscillations occurred through the period under consideration.

Used in the thermodynamic sense the concept of stability may only be applied to systems in a true state of equilibrium. In the case of the rotating earth-surface which intermittently receives and emits energy, only molecular associations with relatively great bond energies may be considered to be stable throughout an entire rotation period. If the rate of rotation of the earth were greatly increased to the point where its temperature remained constant, labile molecules would also assume and maintain certain stable molecular states characteristic of the equilibrium at that temperature. However, if the rate of rotation were then to be gradually decreased, the populations of the various labile molecules would begin to fluctuate in a systematic manner.

In a very complex material aggregate (such as the earth's surface) composed of a great number of types of molecules, certain systemic molecular associations are possible in which the amount of the fluctuation in the populations of constituent molecular species would be reduced. While, strictly speaking, from the thermodynamic standpoint, we may not consider associations accomplishing this effect to be in a more stable state than similar non-associated matter, such an association more closely approximates the condition of relatively constant populations of molecular species, which is a stable state. Thus, there may be a tendency of the matter undergoing changes of state accompanying temperature changes to maintain the mean temperature state or to favor associations which more closely approximate this state.⁴

The fluctuation in temperature in itself acts as a "shuffling factor" which increases the probability that favored associations will occur and that non-favored associations will be eliminated.

⁴ The issue raised here is that of how the probability of formation of certain types of physico-chemical associations in a system at constant tem-

The Origin and Nature of Primordial Systems. In liquid types of milieu of the earth where we have reason to believe the origin of living systems occurred, the concentrations of solute molecules were extremely dilute. Under this condition, associative-dissociative type reactions (many biochemical reactions are of this type) which are initiated by a rise or fall in temperature are free to proceed to an end and are not limited to a state characterized by an equilibrium of the processes of reactant and product formation. Let us assume that it is a characteristic of material aggregates that when subjected to oscillating temperature conditions they tend to maintain the mean temperature state or to favor any associative condition which deviates the least from this state. In this type of milieu this tendency could become operative by the following process. *If the reactant molecules undergoing population fluctuations attendant to the temperature oscillation could be maintained in closer (more concentrated) physical association with the product molecules, the fluctuation in the populations of molecular species would be reduced, for the reaction could not proceed to an end because of the establishment of the reaction equilibrium condition.*

From our standpoint, the only significance to be attached to the question of the existence of the tendency we have discussed is whether or not we can regard the existence of such primordial associative systems to have been highly probable under these circumstances.

Since the carbon-nitrogen type molecules are the ones which are readily involved in reactions initiated by and involving relatively small changes in energy, they are the ones which may tend to associate and form such primordial systems. It is just such a factor as this with which we are concerned, one which may favor the formation of systems of molecular systems characterized by a high content of carbon and nitrogen atoms. Such association complexes could be effected in various ways involving

perature is affected by the instituting of temperature oscillations having a mean temperature equal to the previous constant value.

membranes, cohesive and adhesive actions, electrostatic and hydration forces, etc. Traube (1864, 1866, 1867) and Leduc (1907) have described experiments where the formation of membranes takes place in certain simple chemical systems; Bungenberg de Jong (1937) has given a detailed treatment of coacervation leading to the formation of droplets and semi-liquid colloidal gels (see also Langmuir, 1938), and it is well known that molecules of an aggregate which lower the surface tension tend to accumulate at the periphery.

Considering the nature of the processes which could effect a systemic association of the material in question, the systems formed would be many and small rather than few and large. In simple fluid-type systems, differentiated from the environment by membrane type boundaries, the constituents are in thorough physico-chemical association. This is in contrast to grosser systems bounded through the action of other properties of matter; an example of such a system is the earth, the components of which are held together mainly by gravitational attraction. Consequently such associations are able to display material properties (both intra-systemic and with respect to external phenomena) which are not manifested to such a marked extent, when at all, in other states, *viz.*, reaction equilibrium through concentration of reacting molecules, osmosis, colloidal phenomena, etc. Because of the operation of these factors, the systemic reactions to external conditions take on the aspect to a very pronounced degree that their adjustment to a changing environment tends to oppose the impressed effect, such that to a certain degree the systems tend to maintain their identity. Thus, *these systems, by their very nature, may exhibit to a marked degree certain attributes of life, though on a very primitive level.*

In order to illustrate this property let us consider the membrane-type boundary characteristics in greater detail. We have postulated the function of the membrane to be that of concentrating the bounded molecules to prevent associative-dissociative type reactions from proceed-

ing to the reaction oscillation limits they would attain if the concentration were not effected. If the membrane were non-permeable to any materials of the milieu, the concentration of the solute molecules of the milieu could not be changed by the formation of such a membrane about any portion of it. On the other hand, if it were permeable to all the materials of the milieu, it would not even differentiate a system from the surroundings. In general, then, it is only the selectively permeable type of membrane which can become operational in this manner.

Suppose such a membrane which was permeable to the solvent molecules were to form about droplets of solution. Depending upon the circumstances, the membrane might shrink by the action of surface tension forces, all the while expelling (or ceasing to enclose) molecules to which it was permeable. When the concentration of non-diffusing solute molecules increased to a certain point the expulsion of diffusing solvent and solute molecules would necessarily cease. Thus, it is not difficult to postulate a type of mechanism whereby the formation of a selectively permeable membrane might result in the origin of systems of increased concentration of certain solute molecules relative to the dilute environmental milieu from which they were differentiated. A concentration of solute molecules and membrane formation might also be induced by a mere evaporation process of wave-splashed, tidal-formed, etc., droplets or small pools of water. Membrane-type surfaces formed might remain intact when these concentrated droplets returned to the body from which they were separated, for various factors, new or already present, operating in the interval might (because of the greatly modified surface-volume-concentration relations) result in a sufficient alteration in their properties.

It is important to note a significant property of a system containing labile molecules in its relation to stable matter of the environment. The components of the earth's surface consisting only of stable type molecules absorb all the afferent energy through simple tempera-

ture increase, and thus, merely heat up and cool off as day-night periods proceed. On the other hand, components which also contain appreciable numbers of labile molecules absorb a portion of the afferent energy as potential energy through the mechanism of endothermal reactions. The temperature changes of these latter components are, thus, relatively less than those of the former.

Thus, the rotation of the earth which results in the exposure of its surface to an intermittent afference of energy gives rise to two different types of physico-chemical effects among the surface constituents. The first is the simple change of the temperature of aggregates of stable molecules. The second is the occurrence of endothermal-exothermal reaction cycles among labile molecules, aggregates of which for this reason undergo relatively lesser temperature changes. In systems of relatively greater concentrations of labile solute molecules there is a lesser fluctuation in molecular populations of material involved in associative-dissociative type reactions from the mean temperature condition. For this reason we postulate that the formation of such systems may be favored and that it may be a highly probable event under these conditions.

Reactive Properties of Primordial Systems. When a system with membrane-type boundaries is subjected to environmental change its reactive properties are markedly different (as mentioned above) from material in the non-bounded state. For example, suppose such a system were to encounter an environment in which some new substance were present in appreciable concentration and that this substance were diffusible through the membrane of the system. Then, whatever associative-dissociative type reaction this new substance might enter into with intra-systemic non-diffusing molecules which would give rise to non-diffusing product molecules could not proceed to an end, but would reach an equilibrium state as the concentration of the product molecules increased. This is in contrast to the circumstances in the case of the

dilute environment where such reactions would proceed to an end. Thus, such a system has the property that certain types of environmental disturbances will be opposed or counteracted to a certain extent by the system, and by this mechanism the system will tend to maintain itself. Of course, the circumstances of changed environment, the character of the change, the selectivity of the membrane, the nature of the effect on intra-systemic molecules, the nature of the product molecules, etc., impose a complexity on the problem such that we may only treat it in its most elementary aspect.

In view of the properties which a selectively permeable membrane imparts to the intra-systemic constituents it bounds, we can readily understand how the system might change in a great number of ways in response to various changing or new environmental stimuli. The considerations of how the composition and size of these systems might be modified are relatively straightforward, as is the nature of the mechanism (discussed above) whereby they tend to oppose impressed effects under certain conditions. Almost all the effects of the environment which do not destroy living systems outright are such that they can be reduced in the final analysis to an alteration of concentrations of material and equilibrium states of the cell protoplasm. Thus, this systemic bounded nature provides a basis for the "evolution" of these association complexes to states which might be characterized by the properties of life. The progressive effects of varying environmental agents and stimuli upon primitive living systems probably constituted the causal mechanistic evolutionary factor which served to evoke the incorporation of more elaborate mechanisms of maintenance and response.

Reproduction, in its most primitive form, was probably merely a division process wherein two units were derived from one because of the attainment of a state of physical instability. Suppose, for example, that during a certain season or period of the temperature oscillation,

material of the environment to which the systemic membrane was permeable formed non-diffusing products which increased the size of the system to the point that it was physically unstable and divided into two systems. If, under the systemic conditions the reactions were not reversible, say during the alternate season or period of the temperature oscillation, then this process of reproduction through simple fission might take place continuously at certain periods in the seasonal or diurnal cycle. Thus, *in its primitive form reproduction may have been associated with such periodic action and, at any rate, with such a simple process.*

III. FURTHER ASPECTS OF LIFE

The Autotrophic Bacteria. Various investigators (Jordan and Burrows, 1942; Osborn, 1917; Winogradski, 1890; etc.) consider it plausible that of the forms of life known to us the autotrophic bacteria are the most primitive, especially in view of the fact that in this group are the only non-photosynthesizing organisms that can exist independently of other forms of life. These organisms are widely distributed over the surface of the earth and there is geological evidence (Walcott, 1915) of the existence of a similar type of bacteria 30,000,000 years ago. A point of great interest is that the decayed rocks of alpine summits, where no other life exists, were found by Müntz (Osborn, 1917) to be swarming with the ferment of the autotrophic nitrifying bacteria. Osborn points out that this is also true of the limestones and micaceous schists of the Pic du Midi in the Pyrenees, and the decayed calcareous schists of the Faulhorn in the Bernese Oberland. This seems especially interesting because of the great temperature extremes that may exist at great altitudes. From the point of view of the postulates outlined above, the existence of such a simple form of life in an otherwise lifeless environment may be of some significance. Since no other forms of life exist in these localities it may be assumed that such environments

are unfavorable to life. Perhaps the nitrifying bacteria have evolved from primitive living systems which were present at the time of formation of these mountain ranges. Thus, study of the factors involved here might permit reaching some definite conclusions regarding the significance of the presence of the nitrifying bacteria.

Osborn further states that in the arid regions of the earth the nitrifying bacteria do not exist on the dry surface rocks but act vigorously in the soils, and that this action is associated with the need for moisture. While we have postulated that the origin of life results from the evolution of systemic complexes which may have formed under the influence of factors introduced by temperature oscillations, it is by no means true that oscillating temperature conditions favor the continued existence of living organisms. In fact, because of the nature of protoplasmic constituents, we know that, generally, constant temperature is most favorable. The fact that the nitrifying bacteria exist in the lower layers of soil at depths of six to ten feet in arid regions may also be associated with the relatively constant temperature existing at such depths. The nitrifying bacteria of the high-altitude mountainous regions would probably also be found at depths rather than at the surface but for the rocky nature of the lithosphere.

The Relatively Constant Temperature. The fact that living forms have either developed warm-blooded systems or tended to associate themselves with conditions of relatively constant temperature is most significant. The constituents of protoplasm are only stable under highly specific protoplasmic conditions of temperature, pH, concentration, etc. Thus, protoplasm can not maintain its organization and functional activity when these conditions vary beyond certain well-defined points, high temperature limits being especially important. For this reason the evolution of systems to the point where they included protein-type substances could not have proceeded unless mechanisms were simultaneously developed

which maintained the system at relatively constant temperature. Of course, our earlier considerations of the effects of oscillating temperature on material aggregates and our knowledge of the nature of the processes which maintain relatively constant temperature in living forms point to reactions of carbon-nitrogen type molecules as the chief mechanism of this function. The complex protoplasmic molecules are in relatively low entropy states due to their low energy of formation per unit mass, and the protein type molecules are easily denatured by temperature increases. These facts indicate that the feature of relatively constant temperature continued to be closely associated with primordial systems as they evolved to the living state. In this respect we have already pointed out that relatively constant temperature is a characteristic of the material which we postulate to have formed the primordial systems (because a portion of the energy is absorbed as potential energy by this material).

In plants, for example, the solar radiation directly increases the temperature of the plant tissues only to a small extent. This is because almost all the incident radiant energy is used in transpiration, transmitted by the leaves or reradiated. A large portion of the energy retained is converted into potential energy by the photosynthetic reaction. Thus, the magnitude of the temperature oscillation effected in the plant through day-night periods is comparatively small.

The warm-blooded condition was probably developed in certain animal organisms (from the cold-blooded ancestral condition) as a result of their coming to live on the surface of the earth fully exposed to surface temperature oscillations and the vicissitudes of variable climatic conditions. On the other hand, organisms that originated as cold-blooded forms and have remained so are probably those that tended to live in or near water or under other conditions whereby they were not fully exposed to surface temperature oscillations.⁵ Under these conditions the

⁵ With this view in mind, the extinction of reptiles and other forms of life (in prehistoric times) might very readily be explicated as follows: These

need for specific highly adapted temperature regulating mechanisms was not acute, and so the mechanisms that do exist among these forms are comparatively primitive. The factor of *the relatively constant temperature with which living forms are associated thus appears to be a functional characteristic of life, which is inter-related at a most primitive level with the factors which may favor the association of the material systems which we postulate to have evolved to the living state.*

Experimental Work. Very little experimental work has been undertaken from the standpoint of determining the effect of oscillating temperatures upon the basic activities of the simplest forms of life. It has been found, however, that plants are highly sensitive to variations in the natural average of radiation either in a qualitative or quantitative way (Arthur, 1929; Popp, 1926; Schanz, 1919). Spencer and Melroy (1942) have drawn some very interesting conclusions regarding the effects of rhythmic exposures of organisms to various conditions and they believe that further experiments under such conditions are likely to yield interesting and significant results. Richards (1915) has found that the breaking down of the acids through the splitting of the acid compounds in the eacti is a respiratory process caused by the alternate oxidation and deoxidation of the tissues through the action of the sun.

Significance of Protein-type Constituents. *The role of the highly specific protein-type substances of living organisms is probably associated primarily with the*

forms may have become extinct during a period or periods when the atmospheric clouds of water vapor became *markedly* reduced as atmospheric conditions changed. Forms of life of the cold-blooded type, and other types with very primitive mechanisms of constant temperature maintenance, that were not adaptable to a relatively rapid increase of radiant energy and temperature extreme (due to a marked decrease in the blanketing clouds) would not have survived this period. Cowles (1939, 1940 and 1945) has advocated high temperatures and heat-induced sterility as possible factors in the extinction of the late-Cretaceous archosaurs and, in view of the susceptibility of the male germ cells during active spermatogenesis to temperatures above normal for the testis which Cowles (1945) discusses, heat-induced sterility may well have been the actual mechanism.

origin of the functions they carry out, rather than with the primordial origin of the systemic complexes themselves. It is the extraordinary capacity of protein-type compounds, etc., for transformations and changes which lead to more complex states and the transition to the colloid state, and their possession of large quantities of potential energy, which probably led to their association with the living functions. As Oparin has concluded, "Summarizing briefly what has been said we can draw the following conclusions: Hydrocarbon derivatives, such as alcohols, aldehydes, organic acids, amines, amides, etc., undergo important transformations when their aqueous solutions are allowed to stand (often only in sunlight).⁶ In these solutions the dissolved substances undergo reactions of condensation and polymerization, as well as oxidation-reduction reactions; in other words, every type of chemical change occurring in the living cell. As a result, numerous high molecular compounds, similar to those present in living cells, may appear in aqueous solutions of hydrocarbon derivatives on long standing." These reactions occur only under certain conditions such that, while they might easily be expected to occur in such primordial systemic complexes as we have discussed, they would not occur in disorganized complex mixtures of a great variety of organic substances, and in fact, under such conditions the reactions usually take an entirely different course. When such reactions as described above occur in systemic complexes of the membrane-boundary type, the products may become closely associated with the system itself and impart new properties to the complex as a whole. Then, on a more advanced level of organization, these properties would be so intimately bound up with the characteristics of the system that their relational significance would become obscured. Because of this, the significance of protein-type compounds in relation to the living state has probably been misinterpreted.

Present-day Origin. Let us consider briefly the topic

⁶ Parenthetical phrase by author.

of present-day *de novo* origin of life. There is no scientific basis for concluding that life may not be originating continuously upon the earth. The fact that we have no evidence of such *de novo* origin is of no particular significance, for if there is such origin we must anticipate that it would be in units far too small to be treated in the manner in which we are accustomed to dealing with organisms. Furthermore, if life does originate continuously upon the earth, the primordial systems very probably do not take on all the properties of living organisms for some time, until they reach the stage of, say, the autotrophic bacteria. If this is the case, then we should not be aware of the process of origin except in so far as we perceived the product upon a rather complex level. The uniformity of natural phenomena, and their operation under relatively uniform conditions of causality and material state, lead us to favor the view of possible continuous *de novo* origin of life upon the earth, and also uniform expression in the qualities of the living systems formed. It is likely, however, that the changes in the conditions at the earth's surface since the most favorable period for the origin of life have been so great that present-day *de novo* origin, if it occurs, is highly infrequent.

Conclusions. Thus, we have concluded that the basically necessary (though not sufficient) material condition which is required in order that the living state may be attained is an organization upon the level of a system of molecular systems. We have postulated that matter may have reached this level in membrane-type bounded systems as a result of the operation of factors introduced by the oscillating temperatures resulting from the earth's rotation. These, we believe, may favor the formation of association complexes containing labile carbon-nitrogen type molecules in relatively concentrated amounts. As a result of the nature of these complexes their evolution to the point where they display the properties with which we associate life is postulated. These primitive living systems probably evolved to more complex states as a result of reactions and responses to the continually vary-

ing environmental conditions to which they were exposed. The relatively constant temperature of living forms appears to be basically a characteristic of systemic matter of the type we have postulated to have attained the living state. The role of the highly specific protein-type constituents in living forms is probably associated with the origin of their characteristic functions rather than with the basic factors leading to the origin of the primordial systems that have evolved to the living state. The uniformity of natural phenomena and our postulates of the nature of the causal factors in the origin of life tend to support the contention that life may continually arise *de novo*.

By laboratory experiments with cultures of simple living forms exposed to conditions of an intermittent affluence of radiant or kinetic energy, the determination of the importance of these considerations seems feasible. It should also be possible to investigate, from the same standpoint, behavior of suitable material aggregates under such conditions. If these factors are operative in the origin of life, it is reasonable to expect such phenomena to take place upon other planets where similar conditions arise.

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PARTIAL SUPPRESSION OF HAIR DEVELOPMENT INDIRECTLY AFFECTING FRUITFULNESS AND THE PROPORTION OF CROSS-POLLINATION IN A TOMATO MUTANT

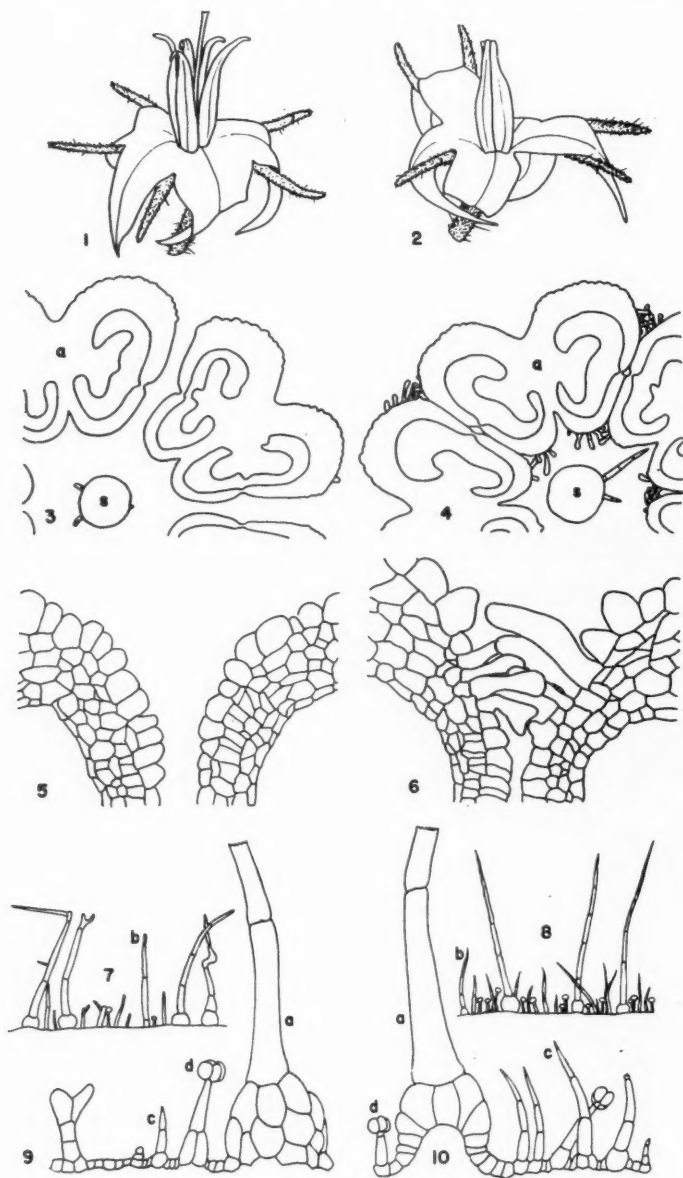
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AN unfruitful tomato plant, designated as No. 12-69, was found in a field of the variety San Marzano near Clarksburg, California, in October, 1944. Measurements of leaf thickness, providing an estimate of chromosome number (Rick, 1945b), indicated that this plant was diploid, an observation verified later in the laboratory by chromosome counts in root-tip cells. Anthers of this plant yielded abundant pollen. The few fruits produced in the field contained about one third as many seeds as fruits of surrounding fertile plants. When self-pollinated by hand, this mutant bore fruits just as seedy as those from normal plants of the same variety. Gametic sterility was therefore not responsible for the unfruitfulness of this plant as it had been found to be for all the unfruitful tomato plants hitherto examined (Rick, 1945a).

The only morphological deviation apparent in the field, aside from unfruitfulness, was the free or dialytic condition of the anthers (Fig. 1). Anthers of fruitful diploid tomatoes are typically connate—that is, they are joined to form a tube surrounding the style (Fig. 2). The profound modification of fruitfulness was tentatively attributed to this slight change in flower form. Other observations, to be described, verify this assumption. Because dialysis means separation of parts normally joined—precisely the condition of anthers in this mutant—plant No. 16-69 and other plants of the same mutant type are called the *dialytic* type; and fruitful diploids with connate anthers, the *normal* type.

¹ Martha O. Rick, the writer's wife, gave much valuable assistance in the course of this study.



MORPHOLOGY AND DEVELOPMENT OF THE DIALYTIC
AND NORMAL TYPES

Low-power magnification of normal-type anthers reveals unicellular epidermal hairs on the anther walls that normally face the style and also on the outer anther walls near the surfaces of contact between the anthers (Fig. 4). Thin transverse sections of living anther cones were cut by means of the freezing microtome. In this material it could be seen that the epidermal hairs of adjacent anthers interlock by weaving between each other and later swelling at the tips (Fig. 6). The hairs are most abundant and the anthers united most securely near their distal ends, this attachment often not existing near their proximal ends. No secretions were observed on these hairs. They do not adhere to a dissecting needle like the multicellular heads of glandular hairs of the tomato stem. Furthermore, pollen grains placed in contact with the hairs do not adhere to them. These epidermal hairs seem, therefore, not to be glandular, nor cemented together in any manner. Anthers of the normal type can be forced apart without injury to the anther except slight damage to the hairs themselves. In this way mature anthers of the normal type are weakly yet effectively bound to each other to form the staminal tube.

Anthers of the dialytic type are devoid of epidermal hairs (Figs. 3, 5). Only the slightest resemblance to the normal type is shown by certain rare cells in the epidermis, which tend to bulge outward somewhat more than adjacent cells (Fig. 3). Thus the mutant anthers are

FIGS. 1 to 10. Camera-lucida outline drawings of plant parts of the dialytic (*dl*) mutant and the normal type. Figs. 1 and 2.—flowers. 2×; Fig. 1.—*dl* type; Fig. 2.—normal type. Figs. 3 and 4.—transverse sections through anthers and style near base of style. 30×; a.—anther; s.—style; Fig. 3.—*dl* type; Fig. 4.—normal type. Figs. 5 and 6.—transverse section through anther walls in outer contiguous region. 125×; Fig. 5.—*dl* type; Fig. 6.—normal type. Figs. 7 and 8.—cauline hairs 5 cm from apical meristem. 10×; a.—long trichomes with multicellular bases; b.—trichomes of intermediate length lacking compound bases; c.—short hairs; d.—glandular hairs; Fig. 7.—*dl* type; Fig. 8.—normal type. Figs. 9 and 10.—cauline hairs. 50×; Fig. 9.—*dl* type; Fig. 10.—normal type.

dialytic because the hairs that normally bind anthers together fail to develop.

The normal type in the variety San Marzano is characterized by a few multicellular hairs near the base of the style (Fig. 4). Still fewer and much shorter styler hairs appear in the dialytic type (Fig. 3).

Anthers of both types start development as separate organs and remain independent until they attain a length of 3 mm. From this time until maturity, anthers of the dialytic type remain free of epidermal hairs. In the contrasting normal type, the hairs protrude earliest in 3.5-mm anthers. The first binding of anthers follows slightly later, in anthers about 4 mm long. The hairs first appear and the anthers first unite at the distal end of the anthers; thereafter these processes develop in a basal direction. By the time the anther is 5.5 mm long, the anther edges are completely populated with hairs, but the anthers are joined by the hairs for a distance of only 1.5 to 2.5 mm from their tips. The hairs continue to grow and bind the anthers until anthesis, when the 8-to-9-mm anthers are usually united for two thirds of their length.

A similar situation in hair development and joining of parts is found in the corolla segment tips. The tips of normal flowers are bound until anthesis by intermeshed multicellular hairs that line the margins of the tips. At anthesis the weak binding effected by these hairs is broken by tension at the tips as the corolla expands. Hairs are developed to a much smaller extent on the corolla segment tips of the dialytic flowers. Hair development is so reduced here that the tips are never united at any stage, but—even in small buds—tend to diverge from each other.

Differences in the development of epidermal hairs are also apparent in most of the vegetative parts. The hairs of the young stem are representative and include four readily distinguishable types, in near agreement with Luckwill's (1943) findings. These appear as follows:

(1) long slender trichomes with bulbous multicellular bases, which are often pigmented with dark purplish anthocyanins (labelled *a* in Figs. 7-10); (2) trichomes of intermediate length without compound bases (*b* in Figs. 7-10); (3) short hairs consisting of only two to five cells (*c* in Figs. 7-10); and (4) glandular hairs, about as long as type *c*, consisting of a short stock of two or three cells surmounted by a tetrad of oblong cells that are responsible for the secretion (*d* in Figs. 7-10). Luckwill's type VII, a very small glandular hair, said to appear sparsely in *L. esculentum*, was not found in this material.

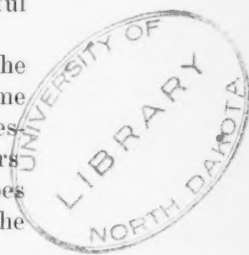
In dialytic plants, these hair types are modified in the following manner: the longer trichomes (*a*) and the somewhat shorter ones lacking compound bases (*b*) are about as frequent as in the normal type, but are shorter and are forked and bent in various grotesque shapes; the short hairs (*c*) and glandular hairs (*d*) are scarcer and somewhat shorter than in the normal type (Figs. 8, 10).

Under proper lighting conditions, the larger trichomes of the two types can be distinguished without magnification. This partial suppression of hair development is systemic, and seedlings of the dialytic type can be identified by the time they have produced two or three true leaves. The dialytic mutant can therefore be added to the dwarfs, the potato-leaf type, the anthocyanin deficiencies, and chlorophyll deficiencies, in the list of useful genetic seedling characters of the tomato.

Considering the syndrome of modifications in the dialytic mutant, it is remarkable that the most extreme effect on hair development—namely, complete suppression—occurs in the hairs that normally bind the anthers. An effect comparable with that observed on other types of hairs might not lead to such striking changes in the fertility of this mutant.

INHERITANCE

The original dialytic plant, No. 12-69, was self-pollinated to produce a family of 19 plants, all dialytic. When



outcrossed to the normal type, No. 12-69 produced entirely normal-type F_1 's. Artificial self-pollinations were made on one of these F_1 plants, yielding three F_2 families, each derived from a single fruit (Nos. 186, 187 and 220 in Table 1). The additional F_2 families in Table 1 (Nos.

TABLE 1
SECOND-GENERATION SEGREGATIONS FOR NORMAL AND DIALYTIC TYPES

Family No.	Normal		Dialytic		d.f.	χ^2	P
	Obs.	Exp.	Obs.	Exp.			
43	22	20.25	5	6.75	1	0.605	0.3 -0.5
44	33	31.5	9	10.5	1	0.286	0.5 -0.7
46	27	21.75	2	7.25	1	5.069	0.02-0.05
186	17	18	7	6	1	0.222	0.5 -0.7
187	25	25.5	9	8.5	1	0.039	0.8 -0.9
220	14	15	6	5	1	0.267	0.5 -0.7
Total					6	6.488	0.3 -0.5
Deviation	138	132	38	44	1	1.001	0.2 -0.3
Heterogeneity					5	5.397	0.2 -0.3

43, 44 and 46) were obtained by open-pollination from other F_1 plants. The observed segregation for dialytic and normal types in these six families is compared, by means of the χ^2 test, with the 3:1 segregation expected of a single recessive gene. A close fit is observed in all families except No. 46. No interfamily heterogeneity is indicated. Possibly the excess of normal plants in No. 46 represents contaminating outcrossings to nearby homozygous normal plants.

At the time these progenies were classified, the differences in vegetative hairs had not been discovered. Plants could be classified as to anther type without difficulty except when the anthers of the normal type occasionally became detached. All scorings were therefore based on the previously described differences in epidermal hairs of the anthers as seen under low-power magnification. The number and size of epidermal hairs of anthers, and especially of the style, is variable in the normal phenotypes; but all attempts to distinguish heterozygotes from homozygous dominants were unsuccessful. The evidence is therefore taken to indicate that this condition of partial hair suppression and consequent dialysis of the

anthers is the expression of a completely recessive gene, to which the symbol *dl* is assigned.

When unfruitful plants of the variety San Marzano were being examined again in the harvest season of 1945, another plant, No. 12-165, was found to exhibit the same features as No. 12-69 found the previous year. A cross was made between these two, and a family of 14 plants grown. Since all plants of this first generation exhibited typical *dl* characters, the responsible genes must be allelic and are probably identical. Whether seed from the same source was used to plant the fields where these two unfruitful plants appeared is not known, but is considered likely, because seed of San Marzano is produced by only one or two firms. Granting this possibility, the *dl* gene could be carried in this stock, hidden in heterozygotes, but expressed in the homozygous *dl dl* segregants—a fact which would account for the two separate appearances. The other possibility—a second mutation at the same locus—must, of course, be admitted.

MODIFICATION OF FRUITFULNESS AND OF THE PROPORTION OF OUTCROSSING IN DIALYTIC PLANTS

The *dl* gene was discovered by virtue of the fact that plants homozygous for this gene are highly unfruitful. Fruit production by the original plants, Nos. 12-69 and 12-165, although known to be low, was not measured; but the fruitfulness of dialytic segregates was observed in the field during the summer of 1945. In an F_2 family, four dialytic plants were surrounded by plants of normal phenotype. Under these conditions, 1.9 to 4.2 per cent. of the flowers of dialytic plants set fruit (Table 2). From 20 to 43 per cent. of the flowers of normal phenotypes in the same family set fruit. A marked difference in fruitfulness of mutant and normal types was also exhibited by another F_2 family. Fruit production of *dl dl* plants is therefore only about 10 per cent. as great as that of their normal sibs. The mean number of seeds per fruit set by open pollination is also reduced in mutant

plants. Total seed production per plant, being the compound of the number of fruits and the number of seeds per fruit, is thus greatly diminished in the dialytic plants.

The degree of fruitfulness of seven male-sterile plants of the variety San Marzano growing in the same area was also measured. Several different genes accounted for the male sterility of these plants. The proportion of flowers that set fruit was 0.7 to 9.4 per cent. Thus dialytic plants have about the same degree of unfruitfulness as male-sterile plants.

TABLE 2
THE RATE OF OUTCROSSING AS MEASURED BY THE PERCENTAGE OF NORMAL PLANTS IN THE PROGENY OF DIALYTIC PLANTS

Clarksburg				Davis				
Female parent	Family no.	Number in family	Percent-normal plants	Female parent	Fruitfulness* per cent.	Family no.	Number in family	Percent-normal plants
12-69	49	14	0	220-1	1.9	38	49	96
	50	4	100			39	36	81
	51	47	21			40	25	48
12-165	57	11	0			41	28	39
	58	14	0	220-5	4.2	42	13	8
	59	20	100			23	34	15
	60	64	17			24	51	57
						25	34	59
						26	40	39
						27	9	89
				220-9	3.7	28	16	81
						29	28	86
						30	36	31
						32	37	22
				220-15	2.5	33	41	17
						34	27	30
						35	37	51
						36	32	13
						37	26	0

* Fruitfulness = Per cent. of flowers that set fruit.

Since these genetically male-sterile plants produce no viable pollen, they can yield fruit only if pollen is transferred to their flowers from nearby fertile plants. (Only fruits containing seeds are considered here; parthenocarp is very rare under these conditions.) Fruit and seed formation by genetically male-sterile plants is therefore a measure of the amount of natural cross-pollination—that is, the amount of functioning pollen transferred from the flowers of one plant to the flowers of another. Since dialytic plants produce about the same

numbers of fruits and seeds as male-sterile plants, it seems likely that the dialytic plants also set fruit only as the result of pollen transfer from flower to flower.

The dialytic character itself can be used to measure the rate of natural cross-pollination—that is, the proportion of all the functioning pollen that consists of pollen transferred from the flowers of one plant to the flowers of another. Since the *dl* gene is recessive, plants with connate anthers (or normal hair type) in the progeny of *dl dl* plants must be hybrids with the normal type. This test was applied to the progeny of the four dialytic plants already mentioned. The following plants were likely sources of pollen in this experiment: (1) the four dialytic plants themselves produced only *dl* pollen; (2) normal plants of *dl +* and *++* genotypes from the same F_2 were alternated in the same row with the dialytic plants; and (3) the flanking rows of plants and all other plants in the field were genotypically *++*. Thus most of the pollen available from nearby plants carried the *+* allele. The proportion of normal plants in the progenies would therefore constitute a minimum estimate of the rate of outcrossing. The seeds from each of four or five fruits taken from each of the four *dl dl* plants were grown as separate families.

Seven families grown from seed produced by the original mutants No. 12-69 and 12-165 under similar conditions in fields near Clarksburg, California, were also included in this test. Considering the rarity of the dialytic mutant, it seems unlikely that any appreciable amount of *dl* pollen was available from heterozygotes in these fields. The percentage of normal phenotypes in these seven families would therefore be a direct measure of the rate of outcrossing. Table 2 summarizes the classification of all these progenies.

The minimum estimates of the proportion of outcrossing per plant at Davis vary from 23 to 66 per cent., and the values for outcrossing of the two original mutants at Clarksburg are 22 and 28 per cent. According to the χ^2

test applied to these two sets of data (Table 3), the inter-family variation is much greater than that expected if the numbers of dialytic and normal plants in each family were distributed at random. The estimate of error must therefore be based on the number of families rather than the total number of plants in all families. On the latter basis, the mean minimum outcrossing value for Davis, 44.9 ± 6.9 per cent., does not differ significantly from that for Clarksburg, 34.0 ± 17.6 per cent.

Comparable tests of the outcrossing of the normal phenotype were not made; but, according to other evidence, it must be much lower than the values obtained for dialytic plants. The male-sterile plants already mentioned produced about 7 per cent. as much seed as male-fertile sibs. This value would be the maximum amount of outcrossing expected of a male-fertile plant, because such a plant produces its own pollen in great quantities, which would compete with the small quantity brought from surrounding plants. The proportion of outcrossing of normal plants at Davis probably falls closer to the $\frac{1}{2}$ to 4 per cent. level obtained at other stations (Currence and Jenkins, 1942; Jones, 1916; Kakizaki, 1929; Lesley, 1924; and Myers and Lewis, 1930).

The percentage of outcrossing to surrounding plants is therefore significantly higher for the dialytic type than for normal tomatoes. Under conditions of the test at Davis, nearly 50 per cent. of the progeny of *dl dl* plants issued from outcrossing to normal plants. The remaining half represent outcrosses to other *dl dl* plants and to *dl +* plants and probably also to self-pollinations. Mechanical self-pollination as in normal tomato flowers could presumably occur. It seems more likely, however, that self-pollination results from transfer of pollen from flower to flower on the same plant, because, as already mentioned, dialytic plants set no more fruit than male-sterile ones.

Even though the *proportion* of cross-pollination is greatly modified in the dialytic mutant, the *amount* of

cross-pollination is probably not much affected. Although pollen brought from other flowers meets little competition in the dialytic flower, the total amount of cross-pollination and total seed production is still low. The amount of self-pollination, on the other hand, is diminished to a tremendous extent by the disruption of the device that ensures self-pollination in normal flowers. Thus the great reduction in amount of self-pollination largely accounts for the increased proportion of cross-pollination.

It seems remarkable that so slight a change in the morphology of the flower could so profoundly alter the amount of self-pollination, notwithstanding the plant's ability to produce viable, self-compatible pollen. In the normal flower, the anthers are united in a tube from which pollen can escape only at the distal end. The anthers dehisce by longitudinal slits in the walls of contact between anthers (Fig. 4). Pollen is thus released into the narrow channels between anthers. Near the sterile anther tips, the pollen moves from these channels into the central space surrounding the style, where it sifts by the stigma, to which some of the grains adhere. Some pollen may also move directly into the central channel from the thecae through ruptures in the ventral binding of the anthers. In the dialytic flower, whose anthers are not bound together, pollen is not directed in any manner after release from the anther; and apparently only by rare chance can it reach the stigma of the same flower. These facts demonstrate conclusively that, at least under our conditions, the binding of anthers by epidermal hairs is essential to normal self-pollination and consequent fruitfulness of the tomato.

Other changes in morphology of the tomato flower—for example, an exserted style—may affect the rate of out-crossing. Lesley (1924) discovered 4.90 per cent. out-crossing in the tomato variety Magnus in contrast to 0.59 per cent. in Dwarf Champion. Styles of the former variety protrude an average of 1.4 mm beyond the anther

tips, whereas those of the latter variety terminate in a stigmatic tip about one mm below the anther tips.

The χ^2 obtained for total variability between families in Table 2 is more than five times as high as that required at the 1 per cent. level of significance in the Davis material and more than four times as high in the Clarksburg material. If, as in Table 3, the Davis data are resolved into the variability contributed by the four parent plants and the variability between families from a single plant, it can be seen that the parent plants in the Davis group differ in the proportion of outcrossing. No evidence at hand would decide whether this significant deviation represents a genetic difference between plants or a difference in insect activity at the different plant locations.

According to Table 3, the χ^2 representing interfamilial variation (indicated as "family"), which is independent

TABLE 3
 χ^2 ANALYSIS OF DATA IN TABLE 2

Locality	Classification	d.f.	χ^2 obtained	χ^2 expected for $P = 0.01$
Clarksburg	Parent	2	1.01	9.21
	Family	5	84.44	15.09
	Total	7	85.45	18.47
Davis	Parent	4	59.01	13.28
	Family	15	141.13	30.58
	Total	19	200.14	36.19

of differences between parent plants, is also significantly large, indicating excessive variability in the proportion of normal plants issuing from different fruits.

If the percentage of normal plants is compared with size of family in each of the families listed in Table 2, a striking relation is revealed. Of the 26 families, eight have 20 or fewer seedlings. Without exception these eight smaller families have either less than 20 or more than 80 per cent. normal progeny. The remaining 18 families, comprising over 20 seedlings apiece, have percentages of normal plants ranging from 0 to 100, but concentrated in the range 15 to 60. Thus the smaller

the number of seeds contained in a fruit, the more likely it is to include a great excess of either normal or dialytic plants; the larger the number, the more likely it is to approach an even mixture of the two types. These results could be explained if it were assumed (1) that all fruits were set by dialytic flowers as a result of pollination by insects, (2) that on a single visit the insect delivers a limited amount of pollen, and (3) that the pollen delivered on a single visit usually is of purely *dl* or purely *+* type. The seed lots of smaller size would then be the product of one or two visits and would more likely be purely of one type or the other; and the larger lots, representing repeated pollinations, would more likely be mixtures of the two types. These assumptions would also explain the very high interfamily variability in the progeny of a single plant. According to them, the randomly distributed unit is not the individual seed, but the group of seeds resulting from each separate insect visit. If, for instance, the mean number of seeds produced by each visit were 10 for the Davis material and 20 for the Clarksburg material, the χ^2 's would be reduced to values corresponding approximately to a probability of 0.5. Minimum estimates of the means—those corresponding to a probability of 0.05—are 5.6 seeds for the Davis group and 7.6 for the Clarksburg group.

The first assumption—namely, that all fruits were set by dialytic flowers as a result of pollination by insects, an assumption suggested by previous comparisons of fruit set by male-sterile and dialytic plants—was tested by permitting a *dl dl* plant to grow during the summer season within a large cheesecloth cage, which shielded it from larger insects. Although this plant produced more than 300 flowers during this period, it failed to set any fruit. A corresponding number of flowers on comparable dialytic plants from which insects were not excluded set about 17 fruits. Insect visits are therefore essential to the pollination of dialytic flowers.

The presence of several *dl +* plants in the Davis plant-

ing would modify the third assumption made above. Pollen delivered from these plants should produce a nearly equal mixture of normal and dialytic offspring. It is not surprising to find, therefore, that the tendency of the smaller families to consist exclusively of either normal or dialytic type is not so marked in the Davis progenies as in those from Clarksburg, where heterozygotes probably did not grow in the vicinity of the original dialytic plants.

The third assumption requires that on a single trip the pollen vectors tend to limit their foraging for pollen to single plants. That a large proportion of the seed set by dialytic plants seems to result from pollen transfer from flower to flower of the same plant would also suggest foraging over very limited areas. Supporting evidence is also found in tests of the rate of natural cross-pollination in various arrangements of male-sterile and male-fertile plants (Rick, 1947). The rate was much higher in a design in which the two types were planted only 6 inches apart and were subsequently permitted to intertwine than in other designs in which the two types were spaced more widely.

Several species of native solitary bees have been observed to collect pollen from tomato flowers at Davis. In tests conducted by Hermann (1921) and the writer, the agency of wind in cross-pollinating tomatoes has been disproven. Fruits containing seeds were not produced by flowers that had been emasculated in the bud stage and that had not been later pollinated by hand. Removal of corolla and androecium evidently renders the flower unattractive to insects but should not render it any less receptive to wind-borne pollen. The available evidence therefore points to insects as the sole agents responsible for cross pollination in tomatoes.

PHYLOGENETIC CONSIDERATIONS

The genus *Lycopersicon* is distinguished from the genus *Solanum* by differences in anther characteristics.

Anthers of *Lycopersicon* dehisce by means of longitudinal slits, extend distally into sterile tips, and are connate in a tube surrounding the style. Anthers of *Solanum*, the genus commonly accepted as ancestral to *Lycopersicon*, shed pollen from terminal pores, do not have sterile tips, and are connivent—that is, converge without morphological connection. By inspecting all specimens of these two genera in the herbarium of the University of California at Berkeley, the writer was able to verify these differences. No exceptions were found in eight species of *Lycopersicon*; the anthers of these species are united by interlocking epidermal hairs. Two exceptions were found in 151 species of *Solanum*. Attachment between anthers was found in *S. bahamense* and *S. Dulcamara*. In the former, large spiny stellate trichomes on the inner side of each anther clasp the style, thus holding anthers in contact with each other. In the latter species, the walls of contact of adjacent anthers have grown together to form a single element in which no separation of anthers can be detected. Anthers of this species cannot be split apart as in *Lycopersicon*. Thus, even in the species of *Solanum* that have connate anthers, the mode of union is entirely different from that in *Lycopersicon*.

In respect to union of anthers, the dialytic mutant resembles the *Solanums*, although it still retains the mode of dehiscence and the sterile tips characteristic of *Lycopersicon*.

The following considerations assume, perhaps erroneously, that insects play no more important a role in cross-pollination in the tomato's native haunts than they do here.

Since normal reproduction of the tomato depends so largely upon union of anthers, as demonstrated by the unfruitfulness of the dialytic mutant, conceivably the origin of the genus *Lycopersicon* may have been intimately related to the appearance of the mutation causing attachment of anthers (probably also related to the ap-

pearance of all the hairs that are so characteristic of the *Lycopersicons*). After appearance of this mutation, the plant had attained a means of automatic self-pollination and no longer depended upon transmission of pollen by insects.² Once the plant had developed this device for self-pollination, it could not return to the original dialytic or connivant condition without a sacrifice in reproductive capacity.

The self-pollinating mechanism would have promoted rapid differentiation of the genus *Lycopersicon* from the ancestral form that first acquired anther attachment. Self-pollination would have ensured complete or nearly complete reproductive isolation of the mutant from the parent *Solanum* species, the degree of isolation depending on the effectiveness of the mechanism. Furthermore, self-pollination would also have speeded differentiation by leading to rapid fixation of genes in the homozygous condition.

SUMMARY

The morphology, inheritance and fertility relations of a tomato mutant are described in this paper. This character behaves as a typical recessive gene in respect to its transmission in selfings of the original mutant, to the characteristics of its hybrids with the normal type, and to second-generation segregations.

The fundamental effect of this gene is a partial suppression of hair development. Fruitfulness is secondarily affected—a fact accounting for the discovery of this mutation. Plants of the mutant type were found

² Luckwill (1943) observed that species of Muller's (1940) subgenus *Eriopersicon* have exerted styles, whereas those of the subgenus *Eulycopersicon* (including *L. esculentum*) generally have stigmas included in the anther tube. Exsertion of the style similar to that found in the subgenus *Eriopersicon* has been found in certain strains of *L. esculentum*. In one instance (Currence, 1944) it markedly reduces fruit and seed production. Thus, despite the anther tube, the exserted stigmas and showy corollas of the *Eriopersicons* might conceivably ensure cross-pollination instead of self-pollination. Until the biology of pollination and fruit setting of the *Eriopersicons* is better understood, further speculation here as to the position of these species in the evolution of the genus is unwarranted.

in fields independently on two separate occasions. Since all hybrids between these two original mutants show the same mutant characters, the genes in each must be allelic and probably identical.

In the mutant the hairs of stems, petioles, pedicels and peduncles are modified in form and reduced in number in the manner detailed in the text. Hairs on the corolla segment tips are also reduced, permitting earlier spreading of the tips. The hairs that, in the normal type, bind anthers together to form the staminal tube are absent, permitting the anthers to diverge. This mutant is called the *dialytic* (*dl*) type, because the separation of anthers is the only obvious change in gross morphology.

Under field conditions the dialytic plants produce only about ten per cent. as many fruits as normal plants; when self-pollinated by hand, they manifest a gamete fertility equal to the normal type. The proportion of hybrids in their progeny indicates that nearly half of the seeds set by the dialytic type under open-pollination result from outcrossing, whereas outcrossing occurs in the normal type to the extent of only one or two per cent. The great reduction in set of fruit and seed indicates that this increase in the proportion of hybrid offspring probably reflects a reduction in the amount of self-pollination to a much greater degree than an increase in the amount of cross-pollination.

These changes in fertility relations of the mutant are attributed to the disruption of the normal mechanism for self-pollination wrought by the separation of anthers. Union of anthers is evidently essential to normal self-pollination of the tomato flower under our conditions. Judging from comparisons with fertility relations of male-sterile mutants and from certain aspects of the data pertaining to the rate of outcrossing, the flowers of the dialytic type set fruits only after pollen is transferred to them by insects.

The bearing of these findings on the origin of the genus *Lycopersicon* is briefly discussed.

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THE ROLE OF TEMPERATURE AND NATURAL
SELECTION IN RELATION TO THE VARI-
ATIONS IN THE SIZE OF THE ENG-
LISH SPARROW IN THE
UNITED STATES

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INTRODUCTION

BETWEEN 1851 and 1881 there were several introductions of the English sparrow, *Passer domesticus*, into the United States from Europe. Their ensuing spread and increase in numbers soon led the U. S. Department of Agriculture to study the importance of this addition to our fauna. Barrows (1889) summarized their findings. Most of the introductions were from Great Britain, although a few came from Germany. Many transfers of small groups of sparrows were made from the original sites of introduction to other parts of the country, where they were fed and protected until their numbers increased to the point where they became a nuisance. This fad of establishing new colonies of English sparrows reached its peak by 1880, before which time Barrows lists 80 such recorded transfers. Regardless of the source of introduction, this process soon led to the spread of the bird over most of the United States.

The relative contribution of Continental sparrows as contrasted to those from England will probably never be ascertained. Barrows (1889, table 1, p. 19) lists the sites of the 16 principal original introductions, but he does not distinguish between those coming from Germany or from England. In response to my inquiry regarding this

¹ The author is indebted to the Emory University Research Committee and the Smithsonian Institution for research grants used in the course of this study. Considerable financial aid was also supplied through the Department of Biology, Emory University, and the Department of Zoology and Entomology, Ohio State University, to cover the charges of shipment of live sparrows or museum skins.

matter, J. W. Aldrich, of the U. S. Fish and Wildlife Service, states that "the original records of these introductions are no longer available." The birds themselves can be our only clue. Fifty-eight males collected prior to 1886 in the region from Washington, D.C., through New York City to the southern New England states (population No. 1, Fig. 1) had the mean wing length of

TABLE 1
MEAN WING LENGTH, ITS STANDARD DEVIATION AND THE AVERAGE AMOUNT OF WEAR OF THE WINGS OF THE POPULATIONS OF ENGLISH SPARROWS SHOWN IN FIGURE 1

Pop. No.	Period	Zone	No. of specimens	Males			Females			
				Mean	σ	Ave. amt. of wear	No. of specimens	Mean	σ	Ave. amt. of wear
1	1	4	58	76.51 \pm 1.42		-.02	36	74.43 \pm 1.51		+.16
2	2	4	98	77.47 \pm 1.57		-.65	66	74.77 \pm 1.16		-.32
3	3	4	66	77.71 \pm 1.68		-1.40	36	74.39 \pm 1.52		-.47
4	4	4	24	77.50 \pm 1.06		+.33	12	74.46 \pm 1.36		.00
5	4	5	19	76.47 \pm 1.35		-.05	18	74.86 \pm 1.14		-.80
6	2	3	13	77.58 \pm 1.33		-.40				
7	3	3	15	77.07 \pm 1.66		-.09				
8	4	4	15	76.90 \pm 1.63		-.62				
9	2	5	59	77.31 \pm 1.55		-.22	52	74.81 \pm 1.67		+.46
10	3	5	14	77.79 \pm 1.36		-1.52	14	73.96 \pm 1.65		+.99
11	4	5	73	77.66 \pm 1.48		-.84	46	74.72 \pm 1.28		-.39
12	4	2	22	77.91 \pm 1.14		-.14	13	74.38 \pm 1.04		-.31
13	3	6	16	76.66 \pm 1.95		+.25	13	74.96 \pm 0.72		-.58
14	4	6	14	77.68 \pm 1.64			12	76.50 \pm 1.38		
15	3	5	20	77.55 \pm 1.42		-1.50	24	74.65 \pm 1.07		+.61
16	2	4	14	76.96 \pm 0.96		-.50				
17	3	4	20	77.10 \pm 1.21		-.56	14	74.29 \pm 0.97		-.87
18	4	4	24	77.52 \pm 1.38		-.60	21	74.71 \pm 1.37		
19	4	2	37	77.94 \pm 1.68		-1.21	21	74.48 \pm 1.14		+.82
20	2	2	22	77.18 \pm 1.30		-.92				
21	3	2	110	75.87 \pm 1.61		+.16	103	73.71 \pm 1.30		-.44
22	3	3	16	75.97 \pm 1.53		+.22				
23	3	4	19	76.29 \pm 1.98			16	73.81 \pm 1.63		
24	3	1	25	76.78 \pm 1.29		-.19				

76.51 \pm 1.42 mm. No sample from England during this time was examined, but 21 males collected between 1890 and 1925 had a mean wing length of 74.93 \pm 1.04 mm. The probability of the *t* test of the difference of these two means is less than .001. The criticism of this comparison is that the specimens were not from comparable periods of time. Witherby (1920) gives the mean for English males as somewhat larger, 76.2 mm with a variation of 72-81 mm. Even so it appears that the earliest adequate sample from the United States gives evidence that the specimens which compose it were larger than the English

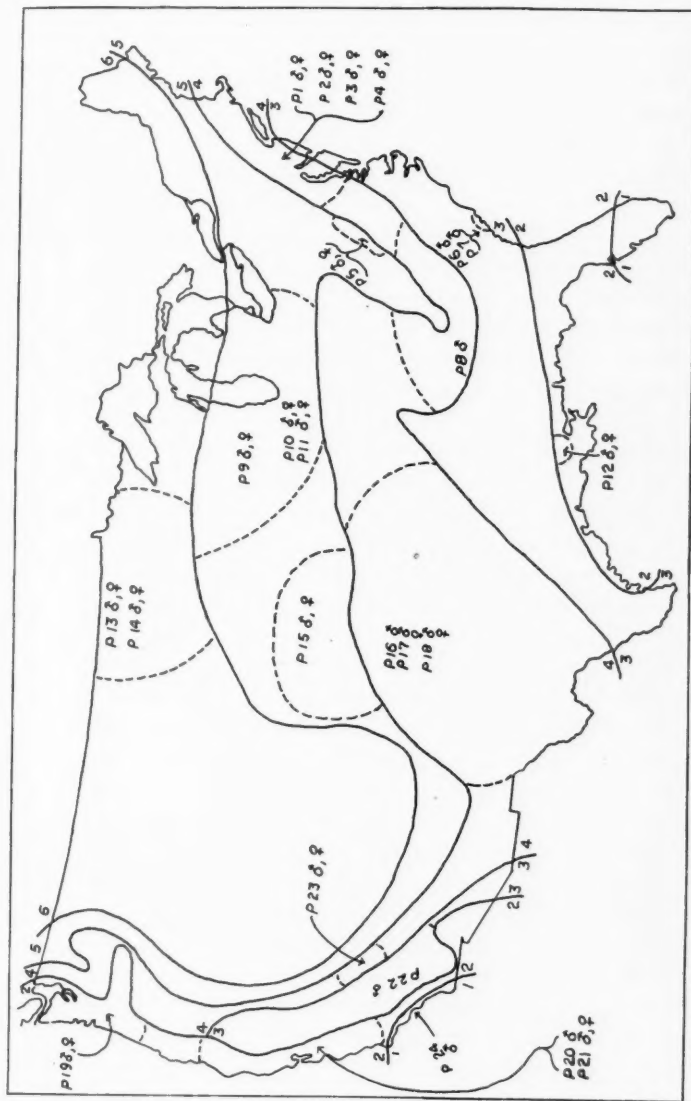


FIG. 1. Location of populations and temperature zones. Solid lines indicate Visser's temperature zones. Broken lines indicate the limits of local populations. A number of specimens utilized in this study are not included in any of these "local" populations. Populations from Period 1 (introduction to 1885): No. 1. Populations from Period 2 (1886 to 1907): Nos. 2, 6, 9, 16, 20. Populations from Period 3 (1908 to 1930): Nos. 3, 7, 10, 13, 15, 17, 21, 22, 23, 24. Populations from Period 4 (1931 to 1945): Nos. 4, 5, 8, 11, 12, 14, 18, 19.

portion of their progenitors. The increase in size during the period to 1886 may be attributed to a selective or a random fluctuation in the heredity of the birds or it may have resulted from the contribution of hereditary material by birds of Continental origin. Witherby gives the mean wing length of 50 such males as 78.9 mm with a range of 75-82 mm.

This problem of the difference between the English and Continental representatives of *Passer domesticus* has received wide attention in the past. On the basis of their smaller size, O. Kleinschmidt (1915) described the English birds as a new subspecies *P. d. hostilis*. On the assumption that the American birds were of English origin, Oberholser (1917) suggested the designation of American sparrows as belonging to the subspecies, *hostilis*. However, the American Ornithologists' Union Committee on Nomenclature (Stone, 1923) ruled against this in the light of Witherby's (1920) contention that *hostilis* was not a valid subspecies on the grounds that less than 20 per cent. of the English birds could be distinguished on the basis of measurement alone. In Fig. 3 it is seen that there may be statistically significant variability within even a 1 mm range. Since this is so the 2.7 mm difference between the means of English and Continental sparrows probably represents a highly significant difference between the heredity of these two groups of *Passer domesticus*. It is just as well that *hostilis* was not considered a valid subspecies because we then would have to designate the American representatives as *Passer domesticus domesticus-hostilis* if we were to attempt to indicate their position. No further investigation of Kleinschmidt's study was possible. He stated that a detailed tabulation would appear later in the journal, *Berajah*, but Hazel Gay of the American Museum Library writes that she is unable to find a record of its publication.

Several extremes of environmental conditions to which the English sparrow became exposed in the United States were greater than normally occur in England or Ger-

many. Included are such places as North Dakota (Cameron, 1907) with its colder winter, the Gulf States with their warmer summers, or as Texas (Montgomery, 1907) and Death Valley, California (Grinnell, 1919), characterized by low humidity.

In invading new and diversified climates it might be expected that some variants of the species might be better adapted to the new conditions than others. This general problem of whether or not there has been any natural selection of new types has interested several biologists during the past five decades. The first important contribution to the problem was that of Bumpus (1898). On February 1, 1898, there occurred a severe storm of snow, rain and sleet in the vicinity of Providence, Rhode Island. One hundred thirty-six English sparrows were found inactivated by the storm and were taken into the Anatomical Laboratory at Brown University. Seventy-two of these birds survived, while sixty-four perished. Bumpus made several morphological measurements of each group. On the basis of his tabulated results, he concluded that the group which perished contained more of those in the extreme size groups—either smaller or larger individuals. To state this in another way, he concluded that the birds of approximately average size could better withstand adverse changes within the environment. On the basis of his present study the writer believes that two of the measurements given by Bumpus may be made with consistent accuracy. These are the measurements for the femur and for the humerus.

Since Bumpus put none of his data to any statistical test of significance, the data for these two bones were re-examined. To better make comparisons with the other data presented in this paper, Bumpus's measurements were transposed from inches to millimeters and grouped into class intervals. Each class interval was assigned as the nearest 0.5 of a millimeter. The birds which perished were compared with those which survived in regard to the numbers falling into extreme and median size

groups. The X^2 values were determined by means of 2×2 tables, and in no instance was any significant difference found. This indicates that extreme size occurred no more frequently among birds which perished than among those which survived.

Although Bumpus's main contention was that those sparrows which survived contained fewer specimens of extreme size, an examination of his tables reveals the fact that those birds which survived were on the average larger (measurements in millimeters):

		Femur		Humerus	
		Males	Females	Males	Females
Survived	18.18	18.14	18.77	18.45
Perished	17.99	18.02	18.53	18.43

Perhaps the real significance of Bumpus's study lies in the larger size of those sparrows which survived. The differences between these means were compared by the t test. It was found that the probability of t was approximately 0.5 for all comparisons except for the humerus among males which lay between 0.1 and 0.05. From such levels of probability we certainly can not attribute any significance to the larger size among those sparrows which survived. However, the occurrence of smaller size among those sparrows which perished in all four of the above measurements indicates that perhaps this was a real difference but that the size of the sample was too small to reveal the significance.

Townsend and Hardy (1909) concluded that English sparrows occurring in New England in 1907 were larger than those found in England in 1907 or in New England during the period of their introduction. The number of specimens used, however, was too few from which to derive any data of statistical significance. Stresemann (1914) tabulates the mean wing lengths (78.4 to 80.0 mm) of several small samples of male Continental sparrows as opposed to the much smaller mean wing length (76.2 mm) of a larger series of sparrows of English origin. He

attributes the larger size of Townsend and Hardy's 1907 New England birds as resulting from their Continental European origin, but this does not explain why New England birds at the time of their introduction were smaller than in 1907. Phillips (1915) approached this problem again with the assistance of a number of collectors over the country, who supplied him with skins. Differences were found between the average wing measurements of birds from various regions, but no tests of significance were applied to the data, nor could he detect any correlation of the geographic distribution of the different size-groups.

A more thorough study of this problem was undertaken by Lack (1940). He determined the average measurements of wing and bill for four groups of males within the United States. The four areas sampled were: (1) Mid-western states: Minnesota, Illinois and Wisconsin, wing 77.6 plus or minus 1.50 mm; (2) Eastern states in the belt from Virginia to Connecticut, wing 77.2 plus or minus 1.59 mm; (3) Berkeley, California, 76.7 plus or minus 1.4 mm; (4) Southern and Lower California, 77.2 plus or minus 1.67 mm. No tests of the significance of the variability of these means were made. According to Visser (1944) these samples as listed come from zones in which conditions associated with severe cold are progressively less severe. On the basis of these means, Lack concluded that there is no significant difference between the samples he selected. Both Huxley (1942) and Mayr (1942) have accepted these results as demonstrating an absence of significant variance between populations of *Passer domesticus*. Huxley suggests that the lack of variability may be due to the short time of their residence in the United States. Lack's method of assembling his data would probably mask any significant differences between populations even if they had arisen. This is because he grouped together specimens collected over long periods of time.

To demonstrate any possible changes in the size of

English sparrows since their introduction one of two procedures must be followed. First, birds collected from successively later periods may be compared. Second, birds from several geographical areas taken during a recent and similar period of time may be compared. These procedures were followed in the present study.

METHODS

Through the cooperation of forty-four² bird-banders scattered over the United States, mostly east of the Rocky Mountains, 611 English sparrows were received, usually alive. To each of these cooperators a shipping cage was sent which was returned to the writer as soon as each had caught a sample, usually of about fifteen birds. The writer is greatly indebted to all these bird-banders for their willing cooperation. Part of these sparrows (263) were made into study skins, and the remainder (348) were made into disarticulated skeletons. Thirty-six³ museums and private collectors were kind enough to make the loan

² Cooperating bird-banders: C. G. Abbott, J. W. Baechle, B. W. Baker, Jno. L. Beal, G. L. Berner, J. T. Birchett, A. E. Borell, G. J. Bristow, J. B. Calhoun, G. D. Chamberlain, L. D. Cool, Jr., R. W. Dexter, D. F. Downey, Mrs. G. A. Dumont, T. L. Engleby, Geo. Fey, Mrs. F. N. Hammerstrom, C. O. Handley, J. W. Haun, C. W. Hibbard, R. D. Hostetter, J. N. Hough, Joe Humphries, Mrs. Wm. Hutchinson, H. E. Jaques, John Lowery, Homer Mumaw, M. K. Nelson, H. F. Perkins, Henry Pluenecke, Mrs. Elizabeth Rapp, F. W. Robl, R. E. Rodeck, Mrs. Leslie Stauber, Mrs. Edna Stevens, T. M. Street, M. T. Sturgeon, Harvey Swanebeck, Mrs. Rowland Thomas, Lillian Thomsen, H. O. Todd, Jr., R. E. Ware, Carl Warren, N. A. Young. Specimens from these sources now in the University of California Museum of Vertebrate Zoology.

³ Museums and private collectors: American Mus. Nat. Hist., Thos. D. Burleigh, Calif. Acad. Sci., Carnegie Mus., Charleston Mus., Chicago Acad. Sci., Chicago Nat. Hist. Mus., Cleveland Mus. Nat. Hist., Colorado College Mus., Colorado Mus. Nat. Hist., Mus. of Comp. Zool., Emory Univ. Mus., Stanley G. Jewett, Univ. Kan. Mus. Nat. Hist., La. State Univ. Mus., Univ. Mich. Mus. Zool., Milwaukee Public Mus., Minn. Mus. Nat. Hist., New England Mus. Nat. Hist., N. Y. State Mus., Newark Museum, Univ. N. C., Museum of Northern Ariz., Univ. Okla., Peabody Mus. Nat. Hist., Acad. Nat. Sci. Philadelphia, Princeton Mus. Zool., Univ. Rochester, San Diego Nat. Hist. Mus., Santa Barbara Mus. Nat. Hist., S. E. Mo. State Teachers College, U. S. Fish and Wildlife Service, U. S. National Museum, M. G. Vaiden, Mus. of Vert. Zool., Univ. Wash. State Mus.

of a total of 1,614 study skins collected in the United States. The writer is also indebted to these sources for their cooperation.

The wing was the only measurement taken from skins. This was taken with the use of a U. S. Bureau of Standards certified millimeter rule to which a right angle joint was attached flush with the zero point of the scale. Wing lengths were recorded to the nearest 0.5 mm. No wings were measured which showed visible amounts of wear, or of which the bird was not adult or was moulting. Despite the fact that all birds with obvious wear to the primaries were discarded there still remained a statistically significant amount of wear between the spring and fall seasons. February, March and April birds were compared with those from October, November and December. Average measurement: 225 "fall" males, 76.87 mm; 397 "spring" males, 76.44 mm; 173 "fall" females, 74.21 mm; 272 "spring" females, 73.86 mm. Probability of the *t* test of the difference of the means for the males was .001, and for the females it was less than .05. It was first attempted to utilize a correction factor based on the above to correct the means of all samples studied until it was called to the writer's attention that the wear for different samples might vary. For this reason each sample was corrected on the basis of the difference shown between the fall and spring portions of that sample. No correction was made unless the fall specimens averaged more than 0.25 mm larger than the spring specimens. Each spring specimen was then corrected to the nearest 0.5 mm before treating the data statistically. In the few cases where the average of the spring portion of the sample was larger than the fall portion, no correction was made. Whereas certain inaccuracies may have been introduced by this method, the writer believes that the corrected wing measurements make it possible to obtain a more accurate representation of the actual situation. The "fall" months were then taken as September through January and the "spring" months as February through July.

In measuring the femur and humerus the above rule was placed on the mechanical stage of a microscope. Using a calibrated ocular micrometer with a 10 X ocular and a 2.8 X objective the lengths of the bones were measured to the nearest 0.01 mm. Although this could be done fairly accurately the measurements were transposed to the nearest 0.1 mm for the purposes of this study. In tabulating the measurements they were grouped in 0.5 mm class intervals.

Three tests were used in determining the significance of the results. These are the t test for comparing two means, the z test for total variance of a population composed of several segregant populations, and the coefficient of correlation, r , between the size of the birds and the temperature zone or time period in which they occurred. The t test for determining the significance of r was made. In each case the probability, p , of z or t was determined by the appropriate tables in Fisher and Yates (1943). Other sources of statistical procedure referred to were Snedecor (1940), Goulden (1939), Paterson (1939) and Fisher (1936).

In the discussion of the results when it is stated that a particular test indicates a significant variance between two populations, a significant variance among several populations or a significant coefficient of correlation, it will be understood that the degree of significance is that of the p listed in the tables accompanying Figs. 2, 3 and 4. Where p is less than .05, .01 or .001 it is meant respectively that in no more than five out of 100 cases, 1 out of 100 cases, or 1 out of 1,000 cases would you obtain such variability or such a high correlation by pure chance. No probability is deemed to indicate significance unless it is 0.05 or less.

Visher (1944) prepared a map delineating six zones in which there is a gradient based on the duration and severity of freezing temperature. Zone 1 is the most moderate and Zone 6 the most severe; it is reproduced in Fig. 1. These zones were used as the basis for dividing the data

on the English sparrow to determine whether or not there was any correlation between the temperature of the environment and the size of the birds. One other division of the data was necessary, that of determining periods of time in which to separate the data. Four periods were decided upon. Period 1 covers the time of their introduction until 1885 by which time they were established in most localities east of the Mississippi River and in a few scattered localities as far west as California. This distribution is mapped by Barrows (1889). Period 2 covers the years 1886 to 1907 during which time the English sparrow had invaded practically all settled regions in the United States. The literature in the *Auk*, Volumes 1 to 30, amply records these invasions. Period 3 covers the years 1908 to 1930, during which time the population of the English sparrow reached a peak and subsequently declined. This phase is discussed by Bergtold (1921) and Eaton (1924), although there is some controversy by others (Anthony, 1921) as to the possibility that the apparent decrease associated with the decrease in numbers of horses was only a relocation of the population with an increase of population in some farming districts. Period 4 covers the years of 1931 to 1944 when it is assumed that the population had reached an adjustment with the established motor vehicle era. At least the literature during this period gives little evidence of any great population change during this time. Weaver (1939b) summarized reports which substantiate the conditions described above for Periods 3 and 4. However, some of his correspondents were of the opinion that the decline during Period 3 was only an apparent one and not a real one due to a shift in the population from urban to rural areas.

RESULTS

During Period 1 the only adequate sample was from Zone 4. This latter population sample was composed of those specimens collected during the time of their intro-

duction. Mean wing lengths for Period 1, Zone 4: 58 males, 76.51 ± 1.42 mm.; 36 females, 74.43 ± 1.51 mm. These were used as the control population with which the means of all other time-zone populations were compared by the *t* test. The 878 males and 620 females occurring during Periods 2, 3 and 4 over Zones 2, 3, 4, 5 and 6 were so distributed as to give adequate samples for all fifteen time-zone populations for the males, and for fourteen time-zone populations for the females; Period 2, Zone 6, was not represented in the comparisons for the females.

The only comparisons where significance was indicated were: the males and females of Period 3, Zone 2, were smaller; the males of Period 2, Zone 5, were larger.

In view of the fact that most of the specimens of both sexes from Period 3, Zone 2, are those which compose the population No. 20 from California (Fig. 1), it might be concluded that the English sparrow in the United States actually is a very stable species, since only one other comparison for the males and none for the females showed any significant variation. Twenty-three males (pop. 20, Fig. 1) collected during Period 2 in the same general region in California, as those mentioned above, had a mean wing length of 77.18 mm; the probability of the *t* comparison with the control was between .1 and .05, whereas with population No. 21, which occurred in the same area during the next period, it was $< .001$. This suggests that the decrease in size of the California birds collected during Period 3 had occurred since their introduction in the San Francisco area (Barrows, 1889, p. 19).

Such a change as occurred among these California sparrows might be due to the action of some selective factor, or it might be due to gene drift occurring by chance (Wright, 1940). Lumping together specimens over an entire temperature zone might mask any changes occurring over more limited areas. For this reason the birds were then divided into local populations. Each population had to meet the following criteria: (a) it must

occur during a single period of time; (b) it must occur in a single temperature zone; (c) no major geographical barrier shall separate the populations into two breeding aggregates. See Fig. 1 and Table 1 for data concerning these populations.

These populations are distributed in two definite types of regions in relation to the variability and intensity of low temperatures as outlined by Visser (1944). There are the peripheral coastal populations (6, 7, 12, 19 to 21, 24) with a relatively mild climate in regard to low temperature. In contrast to these there is the central and northeastern mass of populations (1 to 5, 8 to 11, 13 to 14) which covers an area where there are no major geographical isolating factors between populations and where there is a north to south gradient in severity of cold temperature. The analysis of variance by the z test was applied to each of these groups of populations. For each the probability of z was less than .001. Therefore, we may be fairly certain that in each of these groups of populations the variability expressed by their means is not due to chance.

The variability of the peripheral coastal populations must be due to some other factor than variations in severity of low temperatures since Visser groups all the areas in which they occur into regions of mild temperature in so far as low temperature is concerned. Since it is the purpose of this investigation to study the relationship of the variability of *Passer domesticus* through time and in space to a definite gradient in temperature, no further analysis was made of those peripheral coastal populations.

The first problem which arises is whether there has been a change through time of the mean size of the English sparrow in regions where it has been subjected to periods of low temperature; during which times some size groups might survive better than others. Only three areas contain adequate series through time to enable such an analysis to be made. These are populations 1, 2, 3

and 4, populations 9, 10 and 11, populations 16, 17 and 18. This gave one population from Period 1 and three populations from each of the other following arbitrary periods of time. All specimens from any one period were grouped together to form a single sample. The results are shown in Fig. 2. There is a significant amount of

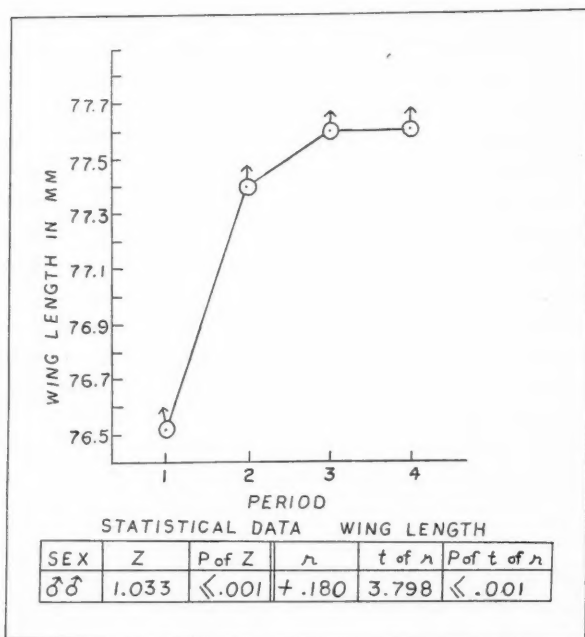


FIG. 2. Gradient in length of wing in relation to the passage of time.

variability among these populations, and as the correlation coefficient shows this variability is so scattered as to give good evidence that there has been an increase in size through time. Note that the most rapid change occurred during the first few decades, then during Period 3 (1908 to 1930) there was much less change, and after 1930 there is no evidence of further increase in size.

The next problem to determine was whether there was

a spatial relationship of this variability associated with the north to south temperature gradient.

This led to a final grouping of the specimens used for an analysis of variability in the English sparrow as evidenced by the measurements of wings. In making this new list only specimens taken east of longitude 105 were used and no specimens taken in peripheral coastal areas were included except those taken in the southern New England and New York-New Jersey area, which were included on the assumption that these areas were so heavily populated by people as to allow a continuous distribution of English sparrows with areas to the west. Only specimens collected after 1908 were included in order that natural selection might have had 40 or more years to act, and because the results of Fig. 2 made it seem probable that little change had occurred after that date. There were only 310 males and 213 females which satisfied these restrictions.

As previously mentioned there is a significant amount of wear from fall to spring. This suggested that those birds shipped alive to the writer might have been exposed to considerable wear of the primaries. As previously, no skins were used in which there was any noticeable wear or in which there was evidence of moulting. Each sex was divided into four groups: (1) museum skins collected during the fall; (2) museum skins collected during the spring; (3) birds shipped alive and prepared into skins during the fall; (4) birds shipped alive and prepared into skins during the spring. The "fall" months were taken as September through January and the "spring" months as February through June. No specimens taken during July or August were used. By using the *t* test on the means of the above groups of specimens it was found that with each sex most differences of mean wing length between seasons were significant. The measurement of each skin was then corrected to the nearest 0.5 mm on the basis of these correction values (Table 2). What this amounted to was correcting each measurement to the

TABLE 2
CORRECTION FACTORS FOR WING MEASUREMENTS IN MILLIMETERS

Source of specimens	Season of collection	Males		Females	
		Zones 3 and 4	Zones 5 and 6	Zones 3 and 4	Zones 5 and 6
Museum Shipped alive	Spring	.71	.97	.29	.33
	Fall	.11	.57	.65	.44
	Spring	.82	1.54	.94	.77

value which on the average would have been expected had the specimen been collected during the fall without having been kept caged prior to its preparation into a study skin. These corrected data on the skins were then reassembled by zones.

The resultant data are graphed in Fig. 3 and the appended table shows that there is a high significance to the

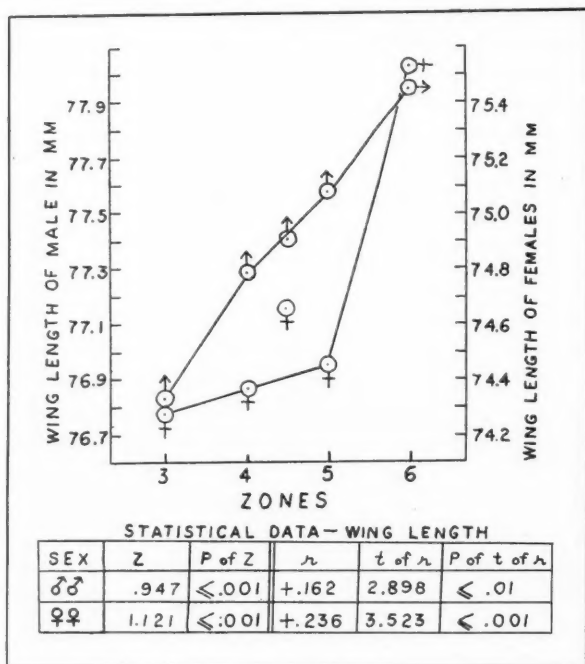


FIG. 3. Gradient in length of wing in relation to temperature. Sex signs between Zones 4 and 5 represent the mean of the means for Zones 3 to 6.

variance among the four samples. The coefficient of correlation data indicates that among this variability there is a high correlation between increase in size and increase in severity and duration of freezing temperatures.

Fortunately a series of skeletons was assembled during the course of this study. However, only 249 of the 348 skeletons were used; these represented 114 males and 135 females. This was because no skeletons were used if there was any indication that ossification was incomplete. The last part of the skeleton to become completely ossified

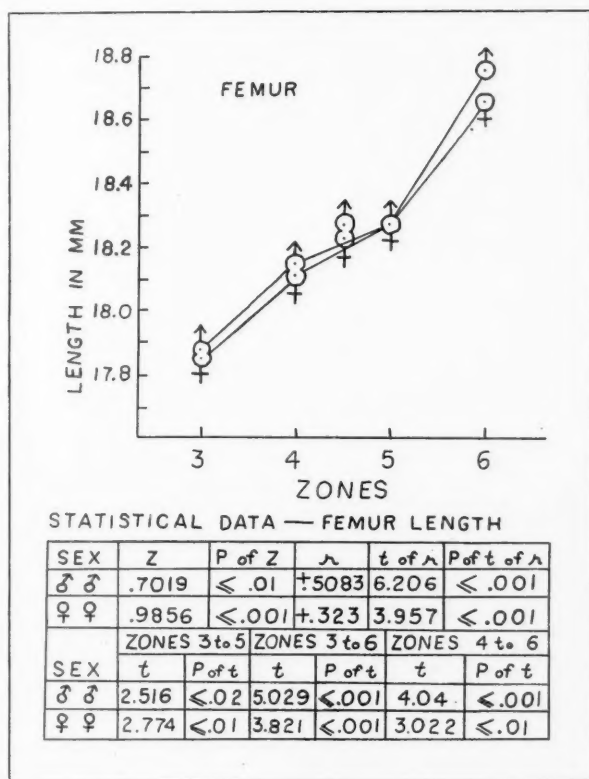


FIG. 4. Gradient in length of femur. Sex signs between Zones 4 and 5 represent the mean of the means for Zones 3 to 6.

is that at the union of the parietals and supraoccipitals. Bones have the advantage over skins in that they are not subject to wear. A number of museums lent skeletons, but these were not used since most of such skeletons were articulated and, therefore, not subject to the previously described method of measuring.

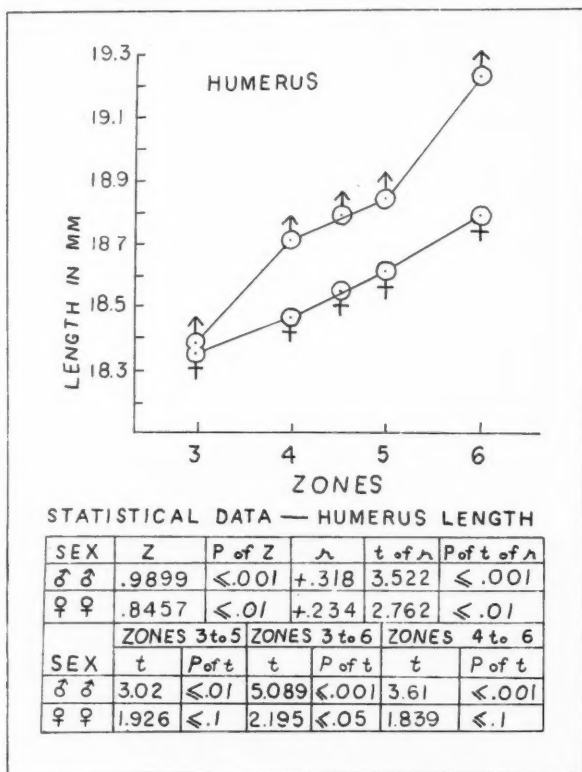


FIG. 5. Gradient in length of humerus. Sex signs between Zones 4 and 5 represent the mean of the means for Zones 3 to 6.

Figs. 4 and 5 with their associated statistical analyses demonstrate even more clearly the close relationship of the size of English sparrows and the degree of severity of cold weather. The fact that these data from skeletal

material conform so closely to that derived from wing measurements arranged by zones (Fig. 3), further supports the validity of utilizing a correction factor for wear used in preparing Figs. 2 and 3.

One population of sparrows in Zone 6 was omitted from the data. This was a population of 17 males and 22 females, mostly from Presque Isle, Maine. The mean measurements for this population were: humerus of males 18.56 mm; femur of males 17.94 mm; humerus of females 18.27 mm; femur of females 17.95 mm. These measurements were found to be significantly smaller than the other birds from Zone 6, those from North Dakota and northern Minnesota, when the means were compared by the *t* test. As it may be seen from Figs. 3 and 4 these birds were smaller than any other group of birds. Here we find an isolated population which varies in the opposite direction from that which might be expected if low temperatures are a selective factor or if increase in body size is directly induced by increased severity of environmental low temperatures.⁴

DISCUSSION

The first problem which arises in the interpretation of the data is to determine whether the increase in size associated with an increase in severity of freezing temperature is directly induced or is due to the differential inheritance of the four samples. Of course it is impossible without breeding experiments to prove this point. However, if the variations are induced directly by the conditions of the environment we would certainly not expect to obtain the high correlation of increase in size with passage of time as is shown in Fig. 2. By direct induction of size variation by changes in temperature the birds of Periods 1 and 2 should have been as large as those of

⁴ The isolation of sparrows in northern Maine is based upon information given me by several bird-banders and other ornithologists in New Hampshire, Vermont and Maine which indicates that through these states there has been a greater reduction of the sparrow population than in any other region of the United States known to the writer.

Periods 3 and 4. Therefore, it is concluded that the change of size through time, and likewise in space, must result from alterations in the heredity of the sparrows.

The second problem which arises is: what is the factor of natural selection that may induce the observed gradient in size? Kendeigh (1944, 1945) has presented much pertinent information relevant to this problem in the English sparrow. From 37° C. to -40° C. there was a straight line increase in metabolism associated with the decrease in temperature. Just the reverse trend occurs concerning loss of weight and survival time without food. Since the body temperature remained constant from plus 37° to minus 40° C. heat loss must have equalled the heat produced by metabolism. He states that "at 25° C. vaporization of water accounts for 16 per cent. of the total heat loss, while at 5° C. it accounts for 9.2 per cent." The assumption is made that below 5° C. vaporization accounts for very little of the loss of heat and that at low temperatures direct radiation from the body is the most important means of heat loss. At low temperatures, below minus 15° C., without food, death usually occurs in less than six hours. Thus, despite the increase in metabolism, heat production can not keep up with heat loss as the food reserves are rapidly used up; the thermo-regulatory mechanism is upset and death shortly ensues. Such being the case we might expect larger sparrows to live longer because their lesser surface in relation to bulk would result in a lessened heat loss by radiation in proportion to the total food reserve.

Since Kendeigh (1944) found no differences of metabolic rate between males and females during the winter it is possible to calculate from his Fig. 1 (1945) the difference in survival time of males and females at various temperatures. Females are nearly invariably smaller than males. Therefore, any differences between sexes should indicate difference due to size.

The following procedure was used in analyzing the data in this figure. Survival times for each 5° C. tem-

perature interval were averaged. The average survival time at two temperature intervals (-40° to 10° C., and 10° to 29° C.) was then determined by averaging these averages. The average survival time from 29° C. to 41° C. was similarly calculated except that the temperature intervals used were 5 , $3\frac{1}{2}$ and $3\frac{1}{2}^{\circ}$ C.; this modification was necessitated by the rapid decrease in survival time at high temperatures.

Males, that is, the larger sparrows, had a survival advantage of 3.66 hours at the lower temperature range and 9.67 hours at the median temperature range. Since survival time at the median temperatures was much longer than at low temperatures it is much more informative to place the advantage in survival time as the percentage of the average survival time for the two sexes. On this basis the larger birds live 24 per cent. longer at low temperatures and 18 per cent. longer at median temperatures. It is hard to escape the conclusion that size is of survival value and that the lower the temperature the greater the survival value of increased size. This is in line with the belief that the increase in size associated with increase in latitude commonly observed in homoiothermal animals (Bergmann's principle, Hesse, Allee and Schmidt, 1937) is due to the survival value of the relatively smaller body surface of larger animals at high latitudes.

However, many sparrows in the United States live in regions (Zone 3, Fig. 1, for example) where the weather is moderate in regard to low temperature, but in which there are lengthy periods of high temperature. What, then, is the relationship of high temperature to survival time? Kendeigh shows that metabolic rate increases rapidly between 37° and 45° C. whereas survival time decreases rapidly. Actually the decrease in survival time begins at 29° C. At 43° C. he calculates that 47 per cent. of the heat of metabolism is lost through evaporation of water from the lungs and air sacs. However, since the normal body temperature (between 0° and 35° C.) is only 41.5° C. much of the heat of metabolism is retained

due to the fact that at 43° C. the higher external temperature prevents heat loss by radiation. Body temperature then rapidly rises as the environmental temperature approaches the lethal level, 46.8° C. The calculations previously referred to indicate that between 29° and 41° C. the smaller females have a survival advantage of 12.33 hours. In terms of the percentage of the average survival time of the two sexes the smaller birds live 30 per cent. longer than the larger ones. This would seem to indicate that smaller birds live longer because of a greater ability to dissipate heat by water loss. There is no indication why this should be correlated with small size. Perhaps enough heat even at high temperatures is lost by direct radiation over their relatively greater body surface to be of survival value.

Having come to the conclusion that the expression of size is due to heredity and that size can be of survival value, the question now arises: Are there any observations of extreme temperatures destroying English sparrows? In the compilation of his monograph on the English sparrow Barrows (1889) received many letters regarding their status over its range at that date. Seventeen of these letters cite instances of great reduction in numbers following severe cold weather which the writers attribute to the effect of low temperatures. Some cite specific instances of finding them frozen to death. These records come from the states of Kentucky, Kansas, Illinois, Ohio, Massachusetts, Minnesota and Wisconsin.

On the basis of observations reported from Wisconsin Barrows is of the opinion that most deaths in the winter result from starvation when food is buried under the snow. Following Kendeigh's previously quoted experiments we might expect only the larger birds to survive under such conditions. Three other observers are quoted by Barrows to the effect that when food is available few sparrows die from the effects of cold weather.

As previously discussed in detail in the introduction Bumpus (1898) recorded mortality directly attributable

to a severe snow, rain and sleet storm in Rhode Island. Whereas the sparrows which perished were not significantly smaller than those which survived, it may well be that the smaller size of those which perished was actually significant but that the numbers of the sample were too small to reveal such significance. If the trend of difference (.25 mm for the humerus, and .19 mm for the femur among males) between those birds which perished and those which survived continued in several exposures to inclement weather it would take the exposure of a maximum of only five generations to produce the significant differences such as are revealed in Figs. 3 and 4 between populations of sparrows taken from 1943 to 1945. In all probability the change would not occur in five years due to the overlapping of generations in time, the fact that all sparrows in the same locality are not equally exposed to the rigors of the weather, that the weather is not equally severe each year over all portions of even the same climatic zone, and that progressively fewer sparrows would die due to exposure to the same conditions of the weather because of the gradual increase of the average size of the members of the population.

The English sparrows occurring through most of the United States east of longitude 105 form a relatively continuous breeding population with only distance isolating the gradual exchange of genes between segments of the population. There is essentially no migration; merely local movement of non-breeding birds with aggregations occurring particularly in areas of abundant food supply (Nichols, 1934; Weaver, 1939a). Over this region selection pressure need be only slightly greater than the rate of exchange of members between adjoining breeding groups to account for the establishment of the gradient in size shown in Figs. 2 to 4. Our data on wing lengths indicate that those English sparrows introduced into this country were smaller than the average size at the present time. Therefore, the spread in extremes of these size gradients must have been established within a mini-

num of 50 to 60 years. These highly significant variances and positive correlations of size with temperature could have become established by a selection pressure only great enough, even in the coldest region (Zone 6), to produce average annual increase of the following magnitude: .03 mm for the wing and .02 mm for the femur and humerus.

No attempt is made to analyze the significant variability between populations of sparrows occurring in different portions of the peripheral coastal region of the United States. Perhaps variations in humidity are responsible. It is also conceivable that at the time when these populations were small during the period of their introduction random shifts in heredity occurred due to gene drift. As these populations increased in size the accidentally induced change in average size of the birds would be maintained indefinitely in the absence of selection due to severe cold weather.

SUMMARY

The first introduced colonies of *Passer domesticus* which became established in the United States were smaller than the average at the present time. The wings of introduced males averaged 76.51 mm, whereas the average at the present is 77.60 mm. Large size is of survival value in extreme cold weather, whereas small size is of survival value in extreme warm weather. This has resulted in the development of a gradient in size in which the sparrows occurring in each of four progressively colder zones are progressively larger. This change has occurred through time. The increase in size occurred prior to 1908, whereas no change has occurred between 1908 and 1945. All conclusions from the data on size gradients is based on analysis of variance and coefficient correlations of high statistical significance. There is just as highly statistically significant variance among peripheral coastal populations as among the more continuous breeding populations in the central and eastern

United States. No analysis was made of the cause of the variance in the former area.

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REVIEWS AND COMMENTS

EDITED BY PROFESSOR CARL L. HUBBS

In these reviews and notices of current biological publications emphasis is given to books and major articles which fall within the special scope of *THE AMERICAN NATURALIST*, in that they deal with the factors of organic evolution. **REVIEWS AND COMMENTS** are meant to include also such general discussions, reports, news items and announcements as may be of wide interest to students of evolution. Except as indicated, all items are prepared by Dr. Carl L. Hubbs, Scripps Institution of Oceanography, University of California, La Jolla, California. All opinions are those of the reviewer.

Human Genetics. By R. RUGGLES GATES. New York, N. Y.: The Macmillan Co., 1946: i-xvi, 1-1518, figs. 1-326, 2 vols. \$15.00.

ONE of the most fruitful fields of biological research in recent decades has been the study of genetics. Well-planned and carefully integrated attacks on the problems of inheritance in various animal and plant species have brought experimental genetics to the status of an exact science. The study of the genetics of men has not had the advantage of the experimental method, nor have many integrated attempts been made to elucidate its problems. Rather the studies of human inheritance have been sporadic, isolated and often undertaken by untrained investigators. Nevertheless an imposing array of papers on this subject has appeared, including many which have been based on sound genetic reasoning.

In Gates's new two-volume set, "Human Genetics," an attempt has been made to bring together this widely scattered literature, to put it into some sort of order and to review it critically. The first two objectives have been reached with marked success; the third often leaves much to be desired. As an example of the thoroughness of the compilation it may be pointed out that, although Gates has listed "only key references," the bibliography contains about 5,600 titles. These appear to be well chosen and fairly representative.

The citations are arranged alphabetically following each chapter. Each citation is listed only once, however,

and not always following the chapter where it is first mentioned. Hence it is sometimes difficult to locate a reference quickly and easily.

Perhaps the most important scientific advance in the study of human genetics has been in methodology. The study of human heredity is essentially an excursion into population genetics. Much work has been done on the analysis and the implications of the proportions of genes and of genotypes in human populations, and on the recognition and understanding of "population ratios" as opposed to "Mendelian ratios." Yet Gates is quite inconsistent in regard to these matters. While discussing modern methods in the early part of the work and here and there throughout its pages, he neglects them at critical points.

For example, in discussing the studies of agglutinogens M and N, where there is indication in some populations of a departure from the equilibrium ratio, Gates states that "In a population with M and N in equilibrium, 25 per cent. M: 50 per cent. and MN: 25 per cent. N would be expected." In a number of places throughout the volumes there is evidence of confusion between correlation and linkage. Correlation between two traits in a randomly breeding population is most likely to be due to the action of a single gene or of a group of genes acting on both characters. It is seldom if ever the result of linkage. Linkage of the genes for two traits results in a correlation only among the offspring of particular families of appropriate genetic constitutions. Moreover, such correlations will be positive in about half the families in which they occur, negative in the other half.

Adequate tests for the occurrence of linkage in man have been formulated. These have not been applied, however, in the various instances in which Gates suggests linkage. Nor is it likely that linkage would be found in many of them, since the suggestions are often based on population correlations.

The treatise opens with a chapter on "General Prin-

ciples of Heredity in Man." This is well written and quite complete. There are a few errors: for example, Table 1 has obviously gone awry. Sex-influenced characters and sex-limited characters are not clearly differentiated.

An interesting and informative chapter on human cytology is followed by a general chapter on linkage. The remainder of the book considers the inheritance of various human traits, system by system. Some entire chapters are devoted to an anomaly or a diathesis. Thus there are chapters on eye color and hair color, colorblindness, eye abnormalities, ear abnormalities, albinism, abnormalities of the skin, hair, nails and teeth, anomalies of the skeleton, metabolic defects and derangements, hemophilia and other blood dyscrasias, the blood groups, allergy, hereditary syndromes, abnormalities of the alimentary canal, sexual and intersexual conditions, twins and twinning, muscular and neuromuscular abnormalities, defects of the nervous system, mental defects, normal mental differences, cancer, constitution, congenital anomalies, stature, and anthropological characters.

The preparation of these volumes required an enormous amount of time and labor in searching the literature and compiling the references. It is not to be wondered at that the discussion is at times less pointed and less critical than might be desired. The bibliography is available for those who care to read the original papers. The index of 90 pages is a readily usable tool. The typography is excellent and the illustrations are good. The volumes will find much use as ready reference works to the rapidly expanding literature. It is to be hoped that they will also serve to stimulate interest in research in this field, to the end that integrated, properly planned programs of investigation in human genetics may be undertaken on a large scale.—LAURENCE H. SNYDER, *Department of Zoology and Entomology, The Ohio State University, Columbus, Ohio.*

Bacterial Chemistry and Physiology. By JOHN ROGER PORTER. Iowa City, Iowa: John Wiley and Sons, Inc., 1946: i-vii, 1-1073, 198 tables, 56 figures, 2 charts. \$12.00.

It has often been claimed that with sufficient knowledge it would be possible to explain life processes upon a basis of chemistry and physico-chemical reactions. While there are still many hiatuses in our knowledge of bacterial physiology, perhaps more is known regarding the vital processes of bacteria than of any other form of life. Dr. Porter has made an important contribution to the literature by summarizing in a succinct, logical and interesting manner most of what is known regarding microbial metabolism and the interrelationships between bacteria and their environment. He has thereby satisfied the need for a good text and reference book on bacterial chemistry and physiology.

By virtue of their ability to attack nearly all kinds of organic matter and to transform certain inorganic substances as well, bacteria and allied microorganisms have a pronounced effect upon the chemical and physico-chemical properties of their environment. In turn, environmental conditions influence the life processes of microorganisms. These matters, together with a consideration of bacterial nutrition and many problems of practical significance, are treated quite thoroughly by the author.

A fundamentally sound foundation for what follows is laid in the first chapter by the discussion from a biological viewpoint of the colloidal state of matter, surface tension phenomena, electrophysiology, electrokinetic phenomena, hydrogen-ion activity, and the theory of pH measurements. The technique of counting bacteria by various procedures and the normal growth cycles or phases of bacteria are discussed in the following chapter. The next chapter elaborates the effects on bacteria of such physical agents as electromagnetic waves, ultrasonic vibrations, agitation, electricity, temperature, pressure, surface tension and filtration. In the long chapter on the effects of chemical agents on bacteria the topics considered include the methods of testing disinfectants, the

dynamics of disinfection processes, bacteriostatic agents, antiseptics and the nature of antibiotics.

Details concerning moisture content, elementary composition, nitrogenous components, carbohydrates, lipids, pigments and vitamin content are given in Chapter 5. The chapter on enzymes summarizes a vast wealth of information on the terminology, preparation, properties, classification, activity, specificity and use of microbial enzymes. One of the best chapters in the book devotes 179 pages to the consideration of the growth requirements of bacteria, growth factors or vitamins, essential amino acids, the use of microorganism to assay vitamins and amino acids, and the interpretation of chemotherapy through nutritional studies of microorganisms.

The final three chapters treat the kinds of carbon and nitrogen compounds metabolized by microorganisms and microbial fermentations, with emphasis on those that are of economic or industrial importance. A comprehensive general index and one of microorganisms add to the usefulness of the book for reference purposes. The extended list of the more recent references, at the end of each chapter, omits most earlier references readily found elsewhere.

Although the subject-matter is restricted primarily to bacteria, frequent mention is made of other organisms in order to stress the fact that the physiological behavior of living matter is the same in both the plant and animal kingdoms. As pointed out by the author, "biologists are now beginning to appreciate the fact that certain vital processes can be studied more easily by using microorganisms, and that the information thus obtained often has direct application to the more complex conditions in higher forms of life." This alone is sufficient reason for recommending the book to all biologists and biochemists who desire to widen their scientific horizons. To bacteriologists it will be an invaluable source of information and references.—CLAUDE E. ZOBELL, *Scripps Institution of Oceanography, University of California, La Jolla, California.*

The Ducks Came Back. The story of "Ducks Unlimited." By S. KIP FARRINGTON, JR. New York: Coward-McCann, Inc., 1945: i-xvi, 1-138, many illus. \$5.00.

THE business depression of the early 1930's coincided with a depression of the duck population; but unlike business during the "roaring 20's," the number of ducks available to hunters in the United States had been declining progressively from the turn of the century or earlier. Much attention has been given to the subject during the past 15 years both by governmental and private game organizations. The Migratory Bird Treaty of 1916 and Act of 1918, and the Migratory Bird Conservation Act of 1929 laid the framework for subsequent establishment of breeding and resting refuges for waterfowl as to water, shelter and food and there have been further restrictions on shooting.

Many ducks breed in Canada, where governmental funds of the United States can not be used. A private foundation, More Game Birds in America, surveyed the situation in both the United States and Canada, and its report, "The 1935 International Wild Duck Census," led in 1937 to organization of "Ducks Unlimited" designed to provide leadership and financial support for improvement of duck nesting areas in the prairie provinces of Canada.

The program in Canada has avoided extensive land purchases, but has included various types of improvement for some 150 suitable nesting areas (euphemistically termed "duck factories" and marked by wooden signs or masonry monuments). Provision of water by dams or ditches and by concentrating or deepening transient pools to hold water throughout the duck breeding season; regulation of spring burning and of summer haying to avoid destroying duck nests and young; campaigns to reduce crows, magpies and jack fish as duck predators; salvage of young ducklings from drought spots; censuses of ducks and of local conditions in spring, summer and autumn; and banding of considerable numbers of ducks to determine subsequent movements are some of the ac-

tivities of local personnel engaged in the work. Many of these activities have contributed measurably to improving the duck population and for these efforts Ducks Unlimited and its membership are to be congratulated.

As to the practices of duck shooters in the United States, the take by hunters and crippling losses are considered by Mr. Farrington to be small factors in holding down duck populations. The author argues for reinstatement of baiting and of shooting over decoys and from batteries and sink boxes—practices that have been outlawed in an effort to distribute more widely the pleasures of wildfowling and to limit the total levy by sports shooters.

This volume is patently a claim for great accomplishments by Ducks Unlimited, with much emphasis in text and by portraits on local officers of this organization in the United States—and an appeal for continued financial support from the pocketbooks of duck hunters. This rosy, one-sided account claims that efforts of Ducks Unlimited during 8 years (1937–1944) “is the conservation miracle of all times” (p. 119), allotting very minor credit to natural and supernatural causes, and practically none to work by the United States Fish and Wildlife Service or the state game and conservation departments.—TRACY I. STORER, *University of California, Davis, California*.

Pure Cultures of Algae, Their Preparation and Maintenance.

By E. G. PRINGSHEIM. Cambridge: The University Press, 1946: i–xii, 1–119. \$1.75.

THIS is a small but concise and informative handbook for all who are interested in laboratory cultures of algae, especially freshwater and planktonic algae. Maintenance of pure algal culture is a concern for not only algologists, but also some physiologists, ecologists, hydrobiologists, morphologists, and even researchers in fish and oyster cultures. In this well-digested booklet of Professor Pringsheim, scientific workers will have the benefit of the experience of an outstanding scientist who has devoted 35 years to intensive research in algal culture, and who

"would like to give all who are interested the opportunity of avoiding my mistakes and of profiting by my experience in this field of biology."

There are eight chapters and a foreword by Professor F. E. Fritsch. After the introductory chapter, there is a chapter on selection and preparation of materials and cultures, one on equipment and media, and two on methods of culture. These are followed by a résumé of the whole process of preparing cultures, a chapter on the care and uses of cultures, and one on the culture of the various taxonomic groups. The bibliography contains 159 items.—C. K. TSENG, *Scripps Institution of Oceanography, University of California, La Jolla, California.*

NOTICES OF NEW BOOKS

Insects of Guam—II. Bernice P. Bishop Museum Bulletin, 189, 1946: i-iii, 1-237, 1 map, 58 figs.—In this second series of reports by specialists on insects collected in Guam, the contribution by ROBERT L. USINGER on Heteroptera of Guam (pp. 11-103, figs. 1-27) stands out by reason of its general biological interest. The section on geographic distribution, preceding one on zonal distribution, carries the following conclusion:

In general, Guam may be said to have a depauperate heteropterous fauna, typical of oceanic islands. It fits into the general Pacific island zoogeographic picture, having a high percentage of specific endemics and very few endemic genera. Of the island groups the hemipterous fauna of which is adequately known, Guam is closest to Samoa and the Philippines. The Guam fauna shows no close affinity with Hawaii, the few species found in common being widespread tropicopolitan types. Judging by the distribution of genera, most of the endemic Heteroptera of Guam came originally from the Oriental region by way of the Philippines.

ACTA HYDROBIOLOGICA ET PROTISTOLOGICA

A new international journal is beginning publication, to cover the fields of hydrobiology and protistology. The managing secretary is Professor P. van Oye, University of Ghent, Belgium. Additional members of the board of editors will include Professor Kaj Berg, University of Copenhagen, Denmark; Professor F. E. Fritsch, University of London, England; Professor William Randolph Taylor, University of Michigan, Ann Arbor, Mich., and Dr. Mrs. N. L. Wibaut-Iseebree-Moens, Amsterdam, Holland.

SHORTER ARTICLES AND DISCUSSION

ON RH GENE FREQUENCIES

THE discovery of the Rh factor and its mode of inheritance by Landsteiner and Wiener (1941) and the subsequent proof by Levine and co-workers (1941) that this factor and its recessive allele are responsible for the disease, erythroblastosis foetalis, raise an interesting theoretical question concerning the gene frequencies of these factors in certain populations. The complex series of multiple alleles, all dominant to rh, recently demonstrated by Wiener and others, does not alter the basic problem. In this discussion the general symbol Rh will be applied to these alleles.

The situation in brief is as follows: Erythroblastosis foetalis, frequently fatal in untreated infants, is due to antigen-antibody reaction when an Rh positive child is born to an Rh negative mother. As Rh negative is a recessive, the matings concerned are always of Rh-Rh or Rh-rh male to rh-rh female and the erythroblastotic child is always heterozygous, Rh-rh. Therefore the death of such a child eliminates one Rh and one rh gene from the gene population. Haldane (1942) has pointed out that if Rh has an initial frequency of more than 50 per cent. and rh of correspondingly less than 50 per cent. in the population this elimination of the two genes in equal numbers should result in a progressive lowering of the frequency of rh until the value approaches a point where it is carried in the population only in heterozygous condition. Obviously if mutation from Rh to rh were occurring in the population somewhat more frequently than the reverse mutation there should be a few rh genes in the population. From what is known of gene mutation rates in general, equilibrium should be reached at a point where the rh gene frequency was quite low.

In the white race, however, the frequencies of these genes as determined by tests of large samples are about 60 per cent. Rh and 40 per cent. rh. The question may be asked as to how rh could have gained this frequency against what appears to be a high negative selection pressure. Almost certainly mutation rate alone can not account for this situation. Haldane has offered the formal explanation that such a situation might have arisen from relatively recent hybridization between two racial stocks,

one of which was homozygous for Rh and the other for rh. It is of interest to consider other factors which may have played some role. The explanation offered here is a tentative one, dealing with the probable breeding structure of human populations in the past. No attempt at a complete mathematical formulation has been made.

Let us assume a human population with a low initial frequency of the rh gene but in which Rh mutates to rh more frequently than the reverse mutation. Mutation pressure alone would account for an initial rise in rh and there would be no negative selection against it until homozygotes began to appear in the population. Random mating in a large human population does not occur. Actually such a population is broken up into smaller sub-populations. This must have been even more true in the past than at present owing to a smaller total population and limited means of migration and intercommunication between sub-populations.

Where the sub-populations were small it should happen many times over that in certain of these, by chance, the frequency of rh would rise to a high level. To take an extreme example, if two couples were to colonize a previously uninhabited area and all four of the individuals happened to be heterozygotes drawn from the general population, then the gene frequencies for this incipient sub-population would be 50 per cent. Rh and 50 per cent rh. Through intermarriage of the children of the two families a sub-population might become established in which the gene frequency of rh was above 50 per cent. before any selective elimination occurred. Inbreeding of such a colony would tend to raise the percentage of rh negative genes as here the selection would be against Rh. Conceivably such a population might reach a condition of homozygosis for rh. Eventual migration from such sub-populations would serve to raise the frequency of rh in the general population.

However, it would not be necessary for rh to attain by chance an initial value of more than 50 per cent. in a sub-population before it would have a selective advantage over Rh. In the past average family size was probably determined by factors other than or in addition to infant mortality; families tended to fill up to a certain average size and the children to reach reproductive age in spite of some infant mortality, whether due to erythroblastosis or other causes. Let us assume a sub-population in

which, by chance, the frequency of rh has risen to 30 per cent. Let us further assume that all heterozygous children born to Rh negative mothers die. Only matings of rh-rh mothers to Rh-Rh or Rh-rh fathers will be concerned in selection. Of the men in this population with 70 per cent. Rh genes 49 per cent. will be expected to be Rh-Rh and 42 per cent. Rh-rh. According to the initial assumption in matings of Rh-Rh men x rh-rh women all children will die. In matings of Rh-rh men x rh-rh women half the children, those which are Rh-rh, will die. Suppose that the average family size is six. In the case of the first families there will be no surviving children; six Rh and six rh genes per family will be eliminated. In the other families in which half of the children die the families will tend to fill up with Rh negative children. Thus, while three Rh and three rh genes will be eliminated by the death of the three Rh-rh infants, six rh genes will be added by the addition of the three Rh negative children which fill up the family. The net result in each of these families will be the elimination of three Rh genes and the addition of three rh genes. Considering the proportionate numbers of the two types of families this amounts to a ratio of 71.5 per cent. Rh genes to 28.5 per cent. rh genes eliminated. Since the initial ratio was 70 per cent. Rh to 30 per cent. rh the elimination will bring about a shift in gene frequencies favoring rh.

Actually not all heterozygous children born to Rh negative mothers are erythroblastotic. This fact favors the chance rise of rh genes in sub-populations beyond the equilibrium point even though it raises that point somewhat. For an average family size of four, with the first Rh-rh child born surviving and the second and subsequent ones dying the equilibrium point will be reached at about 66.7 per cent. Rh and 33.3 per cent. rh. For different average family size and different average number of Rh-rh births before antigen-antibody reaction begins to produce erythroblastosis the equilibrium point will fluctuate but will always remain below the 50 per cent. level for rh.

By the time mutation pressure has raised the frequency of rh to a point where homozygotes begin to appear in the general population, the number of heterozygotes will be sufficiently high so that many small sub-populations will be formed with rh gene frequencies of 30 per cent. or higher. The effective breeding size of many human populations will be much smaller than their numerical size, owing to the presence of individuals with no breed-

ing potential. On the postulate of a family filling up to an average size in spite of some infant mortality, once the frequency of rh reaches, by chance, a value near 35 per cent. in a sub-population selection should favor this gene. Eventual migration from such populations should tend to raise the frequency of rh in the general population. It should be noted that even in sub-populations in which selection acts against rh the selective process will be slowed down by the filling up of certain families with Rh negative children following the death of erythroblastotic infants.

Under a given breeding structure the rise in frequency of rh in the general population might have been in progress. Evolution of economic and social pattern might then have accounted for a shift in population structure such that the present gene frequencies represent an equilibrium. However, and this seems more reasonable, shifts in population structure due to changing social and economic factors may be too rapid to allow the system to reach an equilibrium. Such factors as larger sub-populations, urbanization, increased facility for travel, elimination of clan or tribal taboos on marriage outside the group, and smaller families would all be possible factors involved. The most recent factor, the advance of modern medicine and consequent survival of erythroblastotics, has a dysgenic implication providing recent changes in population pattern had shifted the equilibrium point to where rh was being selected against.

Whether or not the explanation offered can account fully for the present high gene frequency of rh in the white population, the factors of population structure and average family size in spite of infant mortality must have played at least a subsidiary role in the frequencies of the genes at the Rh locus. A further study of human ecology may throw new light on this interesting problem.

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